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OF THE

ELISHA MITCHELL Scientific Society

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The Elisha Mitchell Scientific Society

Founded in 1883 through the efforts of Francis Preston Venable

PAUL E. SHEARIN, President

C. W. HOOKER, Vice-President

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Corresponding Secretary

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Recording Secretary-Treasurer

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JOURNAL
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PROCEEDINGS OF THE FORTY-NINTH ANNUAL MEETING OF THE NORTH CAROLINA ACADEMY OF SCIENCE

The forty-ninth annual meeting of the North Carolina Academy of Science was held at the Woman's College of the University of North Carolina, Greensboro, on May 2 and 3, 1952.

The Academy convened in a general session at 10:00 a.m. on the first day with President W. C. George presiding. At this session a Symposium on Genetics and Contemporary Problems was presented.

The business meeting of the Academy was held at 4:30 p.m., at which the following business was transacted:

The minutes of the 1951 meeting were approved as published in the Journal of the Elisha Mitchell Scientific Society **67**: 163–186.

Report of the Executive Committee

The Executive Committee met in Greensboro on May 1 and 2, 1952. At these meetings the following matters were transacted:

1. The committee recommended the appointment of a temporary membership committee with the vice-president of the Academy as chairman.
2. The committee voted to co-operate with the University of North Carolina Extension Service by offering the services of the Academy membership for making talks, in the name of the Academy, to various high school groups.
3. It was voted to suggest to the High School Committee that it provide someone to attempt to interest newspapers in financing the sending of High School Award winners to the National Science Fair.
4. The committee voted that current membership dues are to be paid by May 15 in order to receive both numbers of the Journal. Members paying dues after that date will receive only the second number of the current volume.
5. The committee voted to recommend to the Academy that the complimentary dinner given by the host institution be discontinued and that there be a substitution of a subscription dinner.
6. The following were elected to membership in the Academy:

Aman, Howard E., Science, Camp Lejeune School
Arnold, Donald S., Chemical Engineering, N. C. State College
Ballance, Leon Gerald, Lake Landing, N. C.
Barger, Benjamin, Psychology, Duke University

Bass, Ralph, Catawba College
Bates, William W., Liggett and Myers Tobacco Company
Bird, Rex L., Wildlife Resources Commission, Hendersonville
Boole, John A. Jr., Botany, University of North Carolina
Borden, Richard W., Duke University Hospital
Bowie, Carol Cotton, Psychology, North Carolina College
Breeland, Samuel G., Entomology, N. C. State College
Broad, A. C., Zoology, Duke University
Burgan, Mary Patricia, Physics, Woman's College, U. N. C.
Burgess, Mary Elmore, General Science, Waynesville High School
Cannon, Mrs. Charles A., Concord, N. C.
Cappel, Edward Dan, Botany, N. C. State College
Clayton, Carlyle N., Plant Pathology, N. C. State College
Cowan, Bessie Mae, Librarian, Mitchell College
Crum, J. U., Mathematics, Western Carolina Teachers College
Cuyler, R. Duncan, Entomology, N. C. State College
Dash, Dorothy T. (Mrs.), Fort Bragg School
Davison, Zeta C., Lexington, N. C.
Derrick, F. Ray, Biology, Appalachian State Teachers College
Fahy, William E., Institute of Fisheries Research, U. N. C., Morehead City
Farquhar, Clyde R., Geology, N. C. State College
Ferebee, Ann Wester, Psychology and Education, Shaw University
Foreman, Charles, Physiology, Duke University School of Medicine
Frost, Jane C. (Mrs.), Biology, Woman's College, U. N. C.
Gaskins, Oliver K., Science, Troy High School
Gast, Robert T., Entomology, N. C. State College
Graff, Paul W., Biology, Appalachian State Teachers College
Grodner, Robert M., Zoology, Duke University
Haesloop, John G., Botany, University of North Carolina
Hankla, Donald J., Wildlife Resources Commission, Burlington
Hendricks, Evelyn Elizabeth, Zoology, University of North Carolina
Hendricks, James R., Parasitology, School of Public Health, U. N. C.
Hinds, Anthony K., Mathematics, Western Carolina Teachers College
Kuder, G. F., Psychology, Duke University
Larsh, John E., Parasitology, School of Public Health, U. N. C.
Laxton, J. M., Science, Kings Creek School
League, Elizabeth Anne, Botany, University of North Carolina
Lilly, Charles J., Science, Mitchell College
Lucas, George B., Plant Pathology, N. C. State College
Lyke, Mazel Bowles (Mrs.), Biology, Albemarle High School
MacLeod, Charles Franklyn, Zoology, N. C. State College
Maki, T. Ewald, Forestry, N. C. State College
Mansueti, Romeo, Chesapeake Biological Laboratory, Solomons, Maryland
Miller, Charles E., Botany, University of North Carolina
Miller, Linda White, Elementary Science, Draper High School
Muldrow, William J., Psychology, State Teachers College, Elizabeth City
Nace, George W., Zoology, Duke University
Nusbaum, Charles Joseph, Plant Pathology, N. C. State College
Paris, Oscar H., Jr., Zoology, University of North Carolina
Pickens, A. L., Zoology, Queens College
Prichard, Elmer C., Botany, University of North Carolina
Reese, Elizabeth M. (Mrs.), Physics and Chemistry, Hickory High School
Rice, Theodore R., Shellfish Laboratory, Beaufort
Richardson, Frank R., Zoology, N. C. State College

Rohde, Henry T., Mathematics, High Point Senior High School
Russ, Mrs. S. T., Biology, Shallotte High School
Scott, H. A., Jr., Charlotte, N. C.
Sox, Jason L., Elon College
Sprague, Victor, Black Mountain College
Stainback, Vivian, Science, Stonesville High School
Streicher, Marjorie M., U. S. Geological Survey, Raleigh
Tamis, Robert Howard, Zoology, Duke University
Tilley, Ethel, Psychology, Meredith College
Tillotson, Rex F., Science, Copeland High School, Dobson
Trimble, Ralph M., Applied Mathematics, University of North Carolina
Ullman, Roy R., Psychology, Lenoir Rhyne College
Vernberg, Frank John, Zoology, Duke University
Wilson, Ruth L. (Mrs.), Biology, Mooresville High School
Wolfe, Frances Lee, Mathematics, Woman's College, U. N. C.

7. The following were reinstated as members in the Academy:

Blalock, Thomas J., Chemistry, N. C. State College
Hackney, Edward June, Liggett and Myers Tobacco Company
Hooker, Charles W., Anatomy, University of North Carolina
James, Ben H., Wildlife Resources Commission, Williamston
Longest, Pauline M. (Mrs.), Science, 71st High School, Fayetteville
Peterson, Walter J., Chemistry, N. C. State College

8. The Secretary reported the following losses in membership:

1. By resignation: 3
2. By non-payment of dues: 47
3. By death the following:
   Ruth Margery Addoms
   Ralph Walton Bost
   William de Berniere MacNider
   Ivan Vaughan Detweiler Shunk
   Roy Frederick Sommer

9. The committee accepted the invitation of Chancellor J. W. Harrelson, North Carolina State College, to meet in Raleigh in 1953.

The above reports and actions of the committee were approved by the Academy.

TREASURER'S REPORT

Financial Statement as of September 22, 1952

Receipts

Checking Account balance (from September 1, 1951)	$288.16
Gifts	
Ornithology Essay Award	20.00
N. C. Section of A. C. S.	10.00
W. C. George	3.00
Dues, 1951	4.00
Dues, 1952	1101.00
Total	1426.16

Expenditures

Journal of Elisha Mitchell Scientific Society	530.50
Stationery, printing, programs	135.70
Secretarial commission	110.50
R. Ruggles Gates	103.00
Secretarial aid	81.00
Office supplies and equipment	79.18
High School Committee	73.55
Postage and stamped envelopes	66.69
High School student awards	60.00
Reprints of proceedings	12.75
Meeting expenses	11.20
Bank charges	7.70
Telephone (long distance calls)	5.93
Rubber check	2.50
Total	1280.20
Bank balance	145.96
	1426.16

Savings Account

Balance (from September 1, 1951)	103.02
Interest	2.06
Total	105.08

Total Assets

Checking account	145.96
Savings account	105.08
U. S. Savings Bonds (maturity value)	300.00
Total	551.04

On the twenty-second day of September, 1952, we, the undersigned, examined this account and found it to be correct.

(Signed) F. W. COOK
W. B. FOX
W. A. REID, *Chairman*

REPORT OF THE RESEARCH GRANTS COMMITTEE

The grant of $96.50 for the past year was given to Dr. Maurice Whittinghill of the University of North Carolina. The grant was wholly expended in partial defrayment of travel expenses involved in the genetic study of harelip-cleft palate and the Laurence-Moon-Biedl Syndrome.

A. D. SHAFTESBURY
W. O. PUCKETT
RUTH M. ADDOMS
GERALD R. MACCARTHY, *Chairman*

THE POTEAT AWARD

The award for this year was given to the best paper presented before the Biochemistry and Physiology Section. The selection was made by the members of the section present at the reading of the papers. The winner for this year is Marjorie Swanson of Bowman Gray School of Medicine for her excellent paper, "Some properties of the ATP-ase associated with the nuclear nucleoproteins of rat liver."

REPORT OF THE CONSERVATION COMMITTEE

Resolution No. 1:

WHEREAS, there has been introduced in the Congress of the United States a bill, H. R. 6784, by Rep. Redden of North Carolina, which would transfer the title to "certain land" from the United States Government in Swain County, North Carolina; this "certain land", though not defined as such, comprises approximately seven square miles of the south part of the Great Smoky National Park as of November, 1949; the author of this bill offers no purpose or reason for this proposal; and the bill is an obvious threat to the State's most valuable natural preserve and embodies an attitude most dangerous to all National Parks;

NOW THEREFORE, BE IT RESOLVED that the North Carolina Academy of Science is firmly opposed to the Redden bill, and that the President of the Academy be directed to bring H. R. 6784 to the attention of the officers of the following organizations, with the aim of securing statements on the bill from these groups:

The North Carolina Wildlife Federation
Western North Carolina Associated Communities
The North Carolina Garden Club
The Wildflower Preservation Society of North Carolina.

BE IT FURTHER RESOLVED that the President of the Academy be directed to send a copy of this resolution to all North Carolina Senators and Representatives in Congress, and to members of the House Committee on Interior and Insular Affairs.

Resolution No. 2:

WHEREAS, the National Association of Biology Teachers has initiated a three year project to increase the effectiveness of conservation teaching in high school biology courses on a nation-wide basis through a grant-in-aid from the American Nature Association; there has been organized a state committee in North Carolina of approximately 100 biology teachers who have agreed to undertake some conservation projects next year under the leadership of Ray Derrick of Appalachian State Teachers College, and Dr. S. E. Duncan of the North Carolina Department of Public Instruction; and these projects to be undertaken are mainly studies and activities requiring information on North Carolina and its resources

not available in standard texts and not included in most college courses taken by biology teachers;

NOW THEREFORE, BE IT RESOLVED that the North Carolina Academy of Science endorse the conservation project of the National Association of Biology Teachers, that the members be encouraged individually to assist these teachers through personal conferences, supplying ideas and information, and conducting field trips, wherever possible; and that the High School Committee of the Academy specifically encourage these and other teachers to exhibit the results of such conservation projects.

Resolution No. 3:

WHEREAS, the North Carolina Academy of Science was instrumental in influencing the passage of legislation prohibiting the commercial use of the Venus' Flytrap, and there will undoubtedly be efforts made by several dealers in the Wilmington area to have this legislation repealed at the next Legislature;

THEREFORE, BE IT RESOLVED that the North Carolina Academy of Science reaffirm its interest in this legislation to the North Carolina Department of Conservation and Development; that the Legislative Committee of the Academy for next year be instructed by the President to be on the alert for bills presented to the next legislature to repeal the present legislation protecting the Venus' Flytrap, and do everything possible to defeat such new bills; and that the new conservation committee be authorized by the Academy to work with the North Carolina Wildflower Preservation Society in exploring the need for legislation to protect other wildflowers being exploited commercially and to prepare an adequate control bill for presentation to the Legislature.

Resolution No. 4:

WHEREAS, the North Carolina Department of Conservation and Development is inaugurating a series of conservation congresses with an initial session in November 1952 of all organizations interested in conservation problems and legislation;

THEREFORE, BE IT RESOLVED that the North Carolina Academy of Science express its interest in such a congress to the director of the North Carolina Department of Conservation and Development and offer the help of the Conservation Committee of the Academy for next year in assisting with the planning and execution of such a congress.

Resolution No. 5:

WHEREAS, the importation of exotic species of animals has become increasingly more prevalent and such introductions, without adequate safeguards, may result in the loss of characteristic native sub-species through hybridization, may cause extensive damage of agricultural crops, and may result in the introduction of both animal and human diseases; specific introductions of *Nutria* (an aquatic rodent from South America), rabbits from Missouri, deer from Wisconsin, and

foxes from neighboring states, have been made recently in the state of North Carolina; the *Nutria* have already begun depredations on agricultural crops; bubonic plague is present in wild rabbit populations in western United States and is spreading eastward as a result of the liberation of these animals in eastern states by sportsmen, and rabies is prevalent in fox populations; the present laws are not adequate to prevent these unregulated importations; and the North Carolina Academy has already gone on record by previous resolution as opposed to such practices which endanger our wildlife populations and human welfare;

THEREFORE, BE IT RESOLVED that the North Carolina Academy of Science reaffirm its opposition to unregulated importations and instruct the Legislative Committee for the new year to take immediate steps to prepare a suitable bill for presentation to the next Legislature; that this committee consult with all agencies involved, particularly the North Carolina Wildlife Resources Commission and the North Carolina Department of Agriculture and the State Board of Health in an attempt to produce a bill which will encompass the interests and concerns of all agencies responsible for such work; and that such a bill be presented at the Conservation Congress in November to solicit the support of other conservation groups.

North Carolina Wild Flower Preservation Society

For the information of the Academy members the Conservation Committee is glad to report that a North Carolina Wild Flower Preservation Society was organized in the spring of last year. A fall field meeting was held on the "Carroll Wild Flower Preserve" near Siler City, and a spring meeting was held at Tryon concurrently with the Academy meeting.

The current executive officers are: Mrs. Herbert P. Smith, President; Miss Rebecca Causey, Secretary and Treasurer. Both officers may be addressed at Liberty, N. C. The annual dues are $1.00.

The Committee expresses the hope that many Academy members may join this society and thus help support this worthy effort to conserve our steadily diminishing native flora.

B. W. WELLS
F. S. BARKALOW
W. A. CHIPMAN
R. J. PRESTON
R. L. WEAVER, *Chairman*

The committee was assisted by E. Gibbes Patton, North Carolina Representative of the Nature Conservancy.

The above resolutions and reports were approved by the Academy.

REPORT OF THE HIGH SCHOOL COMMITTEE

The chairman of this committee was asked to accept the appointment in September, 1951. At the suggestion of the Academy secretary a local committee was appointed.

Discussions at meetings of the committee centered around three topics:
1. The desirability of holding science fairs in each area corresponding to a North Carolina Education Association district, with winning exhibits being sent to the state contest which is administered by the North Carolina Academy of Science. Mr. H. A. Shannon accepted the responsibility for that project.
2. Plans for the classification of exhibits so that they might be placed in definite categories for which prizes are offered.
3. The fact that no prizes were offered which were limited to projects in chemistry whereas two prizes were given by the Carolina Biological Supply Company for biological projects. Mr. J. O. Derrick accepted the responsibility for taking up the matter with Cardinal Products, Inc., of Durham.

As a result of Mr. Derrick's correspondence with Dr. S. Thomas Amore of Cardinal Products, three prizes for chemistry projects were obtained: a first prize of $25.00, a second of $15.00 and a third of $10.00.

At the suggestion of Mr. William S. Edmunds of the North Carolina Forestry Association, the title selected for the Forestry Essay Contest was "The economic value of North Carolina's forest resources." Mrs. E. O. Clarkson of Charlotte, contributor of the first prize in the Ornithology Essay Contest, did not offer a title so the committee selected "Birds as community benefactors."

There was some delay in obtaining a mailing list of the teachers of science, but in December a three-page mimeographed announcement describing the classes of exhibits, suggested projects, titles for essay contests, prizes and general regulations was sent to teachers of science in all the schools included in the list from Mr. Shannon's office. The announcement was also sent to all county and city school superintendents. A total of 893 letters was sent.

In January a follow-up letter was sent to all of the schools, reminding the teachers about the contests and of the possible value of contests for stimulating interest in science, calling their attention to the national shortage of trained scientists. Invitations to membership in the North Carolina Academy of Science were included. There were 670 of these letters.

Essays were to be submitted not later than March 15. A total of 40 essays was received. Fourteen essays from one school arrived a week late, with an incomplete address for return, no certification from the principal of the school, and no bibliographies. These were disqualified by the committee. The remaining 26 essays consisted of 10 entries in the Ornithology Contest and 16 in the Forestry Contest, representing only four high schools. It is suggested by the committee that, in the future, entries for each title be limited to four per school. The work of the committee would be greatly lessened if the high school teacher screens the essays instead of sending the work of a whole class to be read by the committee.

Exhibits were to be listed with the chairman of the committee by April 15. A few entries had been received by that date, others came the week following and some were sent directly to the committee at the Woman's College in Greensboro. A total of thirty-three exhibits was received and the committee wishes to

thank Dr. Charlotte Dawley and her group both for their excellent arrangement of the exhibits and for providing the ballots for voting.

A. D. BOND
J. O. DERRICK
MARY C. HELMS
R. M. HELMS
H. A. SHANNON
CHRISTINE WILTON, *Chairman*

HIGH SCHOOL CONTESTS WINNERS

Forestry Essay Contest

(*Sponsored by the North Carolina Forestry Association*)

"The Economic Value of North Carolina's Forest Resources"
First Prize: Kay Mitchell, Mount Olive High School, Mount Olive
Second Prize: James Buchanan, Claremont Central High School, Hickory
Third Prize: Mattie Ann Cooke, Mount Olive High School, Mount Olive

Ornithology Essay Contest

"Birds as Community Benefactors"
First Prize: Jane Blake, Lumberton High School, Lumberton
Second Prize: Mary Stevenson Britt, Mount Olive High School, Mount Olive
Third Prize: Faye Kennedy, Mount Olive High School, Mount Olive

Exhibits Contests

Group I. Living Plants or Animals (*Sponsored by Carolina Biological Supply Co.*)

First Prize: Bobby Turner and Carl Wall, Morganton High School; Clara Lee McComb, Teacher
"A Collection of Snakes"
Honorable Mention: Billy Foster, Greensboro Senior High School; Jack Luttrell, Teacher
"Orchids"

Group II. Preserved Plants or Animals (*Sponsored by Carolina Biological Supply Co.*)

First Prize: Johnny Green, Morganton High School; Clara Lee McComb, Teacher
"Human Organs and Tissues"

Group III. Models

First Prize: Dwayne Lowder, Albemarle High School; Mazel Bowels Lyke, Teacher
"How Some Animals Solve the Food Problem"

Group IV. Original Projects

First Prize: Teddy Cox, Myers Park High School, Charlotte; Frank Eller, Teacher

"Mineral and Rock Collection"

Honorable Mention: John Brooks, Greenville High School; Mrs. Tripp, Teacher

"Model of a Blast Furnace"

Group V. Chemistry Exhibits (*Sponsored by Cardinal Products, Inc.*)

No first prize given.

Second Prize: Tommy Teague, Greensboro Senior High School; Jack Luttrell, Teacher

"Antifreeze Analysis"

Third Prize: Ronald Locke, Greensboro Senior High School; Jack Luttrell, Teacher

"Home Made Cosmetics"

REPORT ON THE ACADEMY CONFERENCE

Dr. Percival Robertson, The Principia College, Elsah, Illinois, presented a paper on "Financing Publications of State Academies of Science." After summarizing the investigations carried on in 1950 by Dr. Forbes Robertson of the Montana Academy, Dr. Robertson recounted the steps taken to legalize state appropriations for Academy Proceedings in Illinois. The Illinois law discussed reads, "to co-operate with the Illinois State Academy of Science and to publish a suitable number of the results of the investigations and research in the field of natural science to the end that the same may be distributed to the interested public."

Among the suggestions and comments about raising money brought out in the discussions were the following:

1. Academies might apply to the National Science Foundation, which has money for publications.
2. In some states the governor's contingent fund may be available.
3. Increase in dues, with major portion allocated to publication.
4. Establishment of special memberships—life, sustaining, institutional, industrial—at special prices.
5. Advertisements in proceedings, journal or news letter. (It was pointed out that a monthly publication could not be supported in this way.)
6. Selling reprints to authors at a profit.
7. Charges made to exhibitors at annual meetings.
8. Selling extra copies of proceedings to state university libraries to be used by them for exchange.

A paper by Mr. Wayne Taylor (Texas Academy) on "Promotion of the Junior Academy of Science" was presented. Mr. Taylor reported that twenty-one of the thirty-five reporting academies have Junior Academy activity. He outlined the purposes of Junior Academies and emphasized the necessity of having the

right persons to sponsor and guide the work; he stressed the value of the Science Clubs of America, with which he believes every Junior Academy should be affiliated. Among the suggestions and comments made during the discussion were the following:

1. Advantage should be taken of the work done in the grades and this should be followed up in the high school.
2. The simultaneous meeting of junior and senior academies may be unwise, since senior members may wish to and perhaps should attend junior meetings.
3. Twenty-one states now have science talent searches.
4. School boards in some states are unwilling to release students for attendance at science meetings.
5. Every academy would profit if it could receive from the secretary of the Academy Conference a list of junior academies and their officers.
6. Visiting groups which go out from universities and colleges to high schools should include more scientists than is the custom.
7. The lack of science courses in some high schools should be remedied.
8. Many high school teachers lack time and/or enthusiasm to participate in junior academy work; even the facilities for such activity are inadequate in some schools.
9. Junior academy activity should be headed by a high school, rather than a college, teacher.
10. Sponsorship of science fairs is difficult to secure and hinders progress in this direction.
11. Winners of state contests should be helped to attend the National Science Fair.

After this discussion President C. L. Baker of Southwestern College introduced the suggestion that more academy members might be elected fellows of the AAAS, thereby becoming eligible to membership in the Council of the Association. The conditions of election are (1) nomination by any three fellows, (2) nomination by the Administrative Secretary of the AAAS (rarely), (3) nomination by a section secretary after approval of the section committee of the AAAS, and (4) automatically when there is a membership status in an affiliated organization for which a research qualification is required.

The afternoon session opened with a paper by Dr. Frank R. Kern (Pennsylvania Academy) on "Promotion of Research in the State by the State Academy." Dr. Kern mentioned the overlapping of the interests of the more specialized professional societies and the academies, which may act to the disadvantage of the latter. He suggested that state academies are not primarily research institutions, but that all have the purpose among others of promoting research. Many academy members are not in the position to devote much time to research, and though the papers published in academy proceedings may not be as significant as those in the specialist journals, each of them adds to knowledge. Academies should be judged by generations of their activities rather than by one paper or one program. Academies, because they bring together scientists from all fields, should stimulate the development of work on interarea problems. The establish-

ment of better relations between scientist and layman is also important in getting public support for research, and academies can do much of this. In closing, Dr. Kern paid tribute to the scientific work of amateurs, and pointed out that academies can do much to encourage the layman to become interested in investigation.

The next paper was presented by Dr. Wm. Henry Venable (Pennsylvania Academy) of Blaw-Knox Co., Pittsburgh, on "Development of Institutional and Industrial Cooperation." Dr. Venable reported on the results of a questionnaire which he sent to 111 of Pennsylvania's industrial laboratories, and from which he received only 27 replies. Eleven persons who answered had heard of the Pennsylvania Academy; one had participated in an Academy program; practically no interest was shown in Academy Proceedings; thirteen had cooperative research arrangements with educational institutions; six were willing to consider presenting papers to the Academy; some interest was manifested in proposed co-operation with junior academy work. In general, however, this small sample of industrial laboratories was apathetic towards the Academy. Dr. Venable recommended (1) more effective publicity, (2) avoiding competition with other socieites by offering something unique, (3) formation by academies of committees to explore educational opportunities offered by industrial concerns, (4) relating the academy program to industrial as well as to academic interests, and (5) establishment of industrial memberships.

Dr. Howard A. Meyerhoff, Administrative Secretary of the AAAS, presented his remarks on "What the AAAS Expects of the State Academies." Dr. Meyerhoff said the AAAS regards the academies as extensions of the AAAS in their areas. He said that academies can aid the AAAS by (1) appointing delegates to the Academy Conference who will be sure to attend, (2) carrying the results and problems of the Conference back to the academies for discussion, (3) having delegates present and participating in the AAAS Council meetings, (4) relaying the deliberations and actions of the Council to the academies and translating them there to terms applicable to the academies, (5) making better use of the AAAS grants-in-aid, and (6) increasing those grants by helping to get more members of the AAAS. Dr. Meyerhoff also pointed out that the AAAS could do something to help the academies and was very anxious that academies call on the Association. The Association can suggest and, in many cases, provide speakers for annual academy or other meetings. The Arden House Report, just adopted by the Council, suggests the sending of panels of speakers to various parts of the United States. If this suggestion is implemented, academies might be able to take advantage of it. Through its publications, the AAAS can provide better public relations. Dr. Meyerhoff urged academies to send news notes for printing in *Science*. The AAAS offers honorary memberships to one boy and girl annually who are winners of the science talent search in each state, and the same recognition is granted to the boy or girl of highest standing in junior academies, though only three academies take advantage of the latter. He stated that the AAAS is committed to support the Academy Conference and the academies, but needs their guidance to make this support more substantial and effective.

Dr. Percival Robertson of the Illinois Academy was elected president-elect and Dr. Leland H. Taylor of the West Virginia Academy was re-elected secretary.

D. P. COSTELLO

REPORT OF THE JOHN BEWLEY DERIEUX MEMORIAL AWARD COMMITTEE

The Committee feels that this year there should be no award given. We feel that the one manuscript turned in to the secretary (or to any of the committee members) is not quite up to the standards that we believe the donor of the award would fully approve. We therefore recommend that for next year the committee give a longer period of time between the announcement and the deadline.

F. W. LANCASTER
OTTO STUHLMAN, JR.
M. L. BRAUN, *Chairman*

REPORT OF THE RESOLUTIONS COMMITTEE

Be it resolved that the North Carolina Academy of Science express to Chancellor E. K. Graham, the faculty and the students of the Woman's College of the University of North Carolina its appreciation for the hospitality, entertainment, and many courtesies extended to its members at this meeting, and that the especial appreciation of the Academy is due to Dr. A. F. Thiel and the members of the local Committee on Arrangements for their part in making this, its forty-ninth meeting, successful and enjoyable.

Be it further resolved that a copy of this resolution become a part of the minutes of the Academy and that copies be sent to Chancellor Graham and Dr. Thiel.

MARY CAUGHEY HELMS

The above resolution was unanimously approved by the Academy.

Memorials to the following members were read before the Academy: Ruth Margery Addoms, Ralph Walton Bost, William de Berniere MacNider, Ivan Vaughan Detweiler Shunk and Roy Frederick Sommer. Each memorial was approved by a rising vote. That to Dr. MacNider is published elsewhere (Science **115:** 489–490; N. C. Med. Jour. **13:** 165–166). Memorials to Dr. Bost and to Dr. MacNider read before the Elisha Mitchell Scientific Society are published in the Proceedings of the Society, in this issue of the Journal.

RUTH MARGERY ADDOMS

Ruth Margery Addoms, Professor of Botany at Duke University, died in Brooklyn, New York, on August 29, 1951.

Born in Haworth, New Jersey, she was educated at Wellesley College and the University of Wisconsin. Before coming to Duke University in 1930, she held teaching posts in Valparaiso, Indiana, at Wellesley College and at the University of Wisconsin. The first woman with professorial rank in the Department of Botany, she not only held a responsible place in her own department, but was

highly respected throughout the University for her scholarliness and dynamic leadership. She was a member of Phi Beta Kappa and Sigma Xi. She served for a number of years on the University Committee on Courses of Instruction. She was instrumental in establishing the Alice M. Baldwin Scholarship Fund. Her wise counsel was sought frequently by students and colleagues who recognized her soundness of principles and discriminating judgment.

Her research contributions were in the field of plant physiology and anatomy. Long interested in mineral nutrition, especially as applied to crop plants, her later researches extended to the field of growth substances and physiological anatomy. She made substantial contributions to these fields.

Her contributions to the civic life of Durham were manifold. As a member of the Durham Council of Girl Scouts, as president of the Durham Branch of the American Association of University Women, as chairman of the Durham Committee of the British War Relief Society, as a charter member and twice president of the Durham Altrusa Club, and as an organizer and first chairman of the Interclub Council of Women's Civic Organizations, she was known and admired by hosts of Durham citizens.

Her vigorous personality, her friendly spirit, her love of people, her appreciation of the good things of life, endeared her to all who knew her. She was a teacher by example as well as in fact. Seldom does one find a person who loved life more or who had such varied interests and abilities. Because she held unswervingly to the things she believed were right and true; because of her absolute sincerity and integrity and the high standards she held for herself and others, the influence of her life is indelibly stamped on those who knew her best. In her untimely death the academy has lost a valuable member; her colleagues, a loyal friend.

LEWIS E. ANDERSON

IVAN VAUGHAN DETWEILER SHUNK

Dr. Shunk, Professor of Botany at North Carolina State College, died very suddenly of a heart attack on July 15, 1951. Though his health had not been good in recent years, he met all his teaching obligations up to and through the spring term of last year.

Dr. Shunk was born at Sherrodsville, Ohio, on February 6, 1891. Part of his high school education was gained in Canada, when his parents returned temporarily to their native country. He received the Bachelor's and Master's degrees from West Virginia University and in 1928 Rutgers University awarded him the Ph.D. degree, where his graduate work in soil bacteriology was directed by the renowned Selman A. Waksman. He was a member of both Phi Beta Kappa and Sigma Xi.

Shortly after a period of army service in the first world war, he came to State College in the fall of 1919 as Instructor in Botany and Bacteriology, later being made full Professor in 1943. In this, his first and only appointment, he most faithfully and successfully served his institution for thirty-two years.

In 1917, before coming to Raleigh, he was married to Olivia Rhoades of Belle Vernon, Pennsylvania. She and a daughter, Geraldine, survive him.

Dr. Shunk's colleagues always recognized in him many admirable traits, steadiness of purpose, loyalty to the best interests of his group, and a willingness to co-operate in all constructive activities seldom surpassed among college professionals. But the characteristic which will perhaps be remembered the longest by his intimates, and envied by them, was his remarkable memory. After a ten year interval, upon turning a particular corner in a town on a trip east, he said to the writer, "Do you recall what we were talking about when we turned this corner ten years ago?" And he proceeded to repeat the whole conversation. It was this unusual memory which made Dr. Shunk noted in his Department for ability to teach the widest range of courses. He not only carried elementary and advanced work in Bacteriology, his special field, but, when occasion arose, taught most of the other courses, during the earlier years of his incumbency. He was an excellent teacher, enthusiastic, yet thorough in the presentation of his material and always fair and just in his student relations.

Though teaching was his major concern, Dr. Shunk carried out research work chiefly in the fields of Bacteriology and Ecology. I note that he presented some six papers before the Academy. The writer is personally greatly indebted to him for collaboration in many ecological investigations. His well stored mind was a good one against which to test ideas. His capacity to enjoy the isolated field camps meant much for the prosecution of the projects.

In the death of Dr. I. V. Shunk, N. C. State has lost one of her most capable and loyal professors, and the Academy a devoted member.

B. W. WELLS

ROY FREDERICK SOMMER

Roy F. Sommer died suddenly at his home on Bayside Road, Miami, Florida, August 24, 1951, at the age of 64.

He was born at Wilmont, Ohio, September 20, 1883. In 1913 he received his A.B. degree from Antioch College, and in 1916 his M.A. from Toledo University. He studied at Ohio State University until 1917.

The next year, he went to Mexico City, where he acted as interpreter to the then president of Mexico, President Obregon. He accepted a position to teach in the National University of Mexico, and two years later in the American College. On December 31, 1923, he married Senorita Lola Manzano, of Mexico.

In 1927 Mr. Sommer left Mexico and settled in the vicinity of New York City where he taught first at City College, and later at New York University and at Long Island University.

While he was in New York, for three years he did research work at Columbia University on the gyroscope. At the end of this time he left teaching and entered the research field for the Sperry Gyroscope Corporation, where he carried on many and varied studies. His achievements in this field were widely recognized.

He stayed with industry until 1942. At that time he went to Augusta Military Academy, Georgia, where he remained one year, after which he went to Defiance College in Ohio. He came from Defiance to Pfeiffer Junior College in 1946.

His interest in science was ever alert, young and alive. He was able to awaken

in many students that spirit of inquiry that leads to advanced study and genuine achievement. His endless patience and kindness made him appreciated by students and fellow workers alike. His courteous manner marked him always a born gentleman.

He is survived by his wife and two daughters, Gladys Sommer Silva and Hazel Sommer Hoinville.

We who knew and worked with him feel that his sudden death is a definite loss to us and to the North Carolina Academy of Science.

BESSIE WALLDROFF

REPORT OF THE NOMINATING COMMITTEE

The committee, consisting of A. S. Pearse, Chairman, J. N. Couch and O. C. Bradbury, submitted the following nominations:

President: O. J. Thies, Jr.; Professor Thies declined the nomination and Otto Stuhlman, Jr., University of North Carolina, was nominated from the floor

Vice-President: Reinard Harkema, North Carolina State College

Secretary-Treasurer: John A. Yarbrough, Meredith College (3 years)

Member of the Executive Committee: Eva Galbreath Campbell, Guilford College (3 years)

Members of Research Grants Committee: Lewis E. Anderson, Duke University (2 years), H. E. LeGrand, U. S. Geological Survey (3 years)

The above nominees were unanimously elected by the Academy. This concluded the business and the meeting was adjourned.

The Academy enjoyed the complimentary buffet dinner given by the Woman's College. At 8:15 the evening session was held in the Lecture Room of the Library with Vice-President Otto Stuhlman, Jr., presiding. The 1951 Poteat Award was presented to Mr. Steve Boyce of North Carolina State College. Dr. Christine Wilton announced the winners of the various high school contests. The challenging address of President W. C. George was enjoyed by the membership and guests.

The following officers were elected for the respective sections:

Biochemistry and Physiology: Chairman, C. E. Anderson; Secretary, Marjorie Swanson

Botany: Chairman, A. E. Radford; Secretary, W. B. Fox

Geology: Chairman, S. D. Broadhurst; Secretary, E. W. Berry

Mathematics: Chairman, H. V. Park; Secretary, H. M. Nahikian

Psychology: Chairman, L. D. Cohen; Secretary, Marion Stanland

Wildlife: Chairman, Win Donat; Secretary, W. L. Hamnett

Zoology: Chairman, E. C. Horn; Secretary, C. E. Jenner

The following constitute the personnel of the standing committees:

Executive: Otto Stuhlman, Jr., Reinard Harkema, John A. Yarbrough, E. C. Cocke, W. O. Puckett, Eva G. Campbell

Research Grants: A. D. Shaftesbury, W. O. Puckett, L. E. Anderson, H. E. LeGrand

Dr. Otto Stuhlman, Jr., President of the Academy for 1953, announced the appointment of the following committees:

Auditing: W. A. Reid, Chairman; F. W. Cook, W. B. Fox
Conservation: H. A. Totten, Chairman; F. S. Barkalow, Jr., A. F. Chestnut, W. A. Chipman, R. J. Preston, Jr.
High School: Mary E. Yarbrough, Chairman; B. B. Brandt, Mrs. Eric Copeland, Claire Freeman, Garmon B. Smith, Christine Wilton.
Legislative: F. S. Barkalow, Jr., Chairman, C. G. Bookhout, B. W. Wells
Nominating: W. C. George, Chairman, H. L. Blomquist, Z. P. Metcalf
Resolutions: Dorothy Adkins
Representative on A. A. A. S. Council: John A. Yarbrough
Representative to Academy Conference: John A. Yarbrough
John Bewley Derieux Memorial Award: E. G. Purdom, Chairman, H. D. Crockford, W. E. Speas

The following papers were presented during the meetings. Those marked with an x are abstracted in these proceedings:

Presidential Address: The responsibility of scientists in this era of subverison of the intellect. W. C. George, University of North Carolina

SYMPOSIUM ON GENETICS AND CONTEMPORARY PROBLEMS

Genetics and the improvement of domestic animals. C. H. Bostian, N. C. State College
Genetic principles and plant breeding. S. G. Stephens, N. C. State College
Genetics and disease. C. N. Herndon, Bowman Gray School of Medicine
Genetics, taxonomy and the races of man. R. Ruggles Gates, Professor Emeritus, Kings College, University of London (Published in full in this issue of the Journal)

BIOCHEMISTRY AND PHYSIOLOGY SECTION

x*Biochemical control of caste in the social life of an insect community.* A. L. Pickens, Queens College
x*An optical analysis of the laminated lens of a typical teleostat eye.* Otto Stuhlman, Jr., U. N. C.
x*Serological studies of the blood of the developing chick embryo.* G. W. Nace, Duke
Influence of ovarian hormones on the rat uterus. Maude F. Williams, Woman's College U. N. C.
x*Fluoride and total liver lipid.* C. L. Yarbro, Bradford Miller and C. E. Anderson, U. N. C.
Some properties of the ATP-ase associated with the nuclear nucleoproteins of rat liver. Marjorie Swanson, Bowman Gray
x*The influence of fluoride on whole body retention of radioactive rubidium.* M. K. Berkut and J. C. Andrews, U. N. C.
x*Blood-clotting studies on dogs injected with radio-gold.* J. H. Ferguson, U. N. C.

BOTANY SECTION

The Myxophyceae of Smith Island, North Carolina. E. C. Cocke, Wake Forest

x*Further studies on the motile spores of* Actinoplanes. W. J. Koch and J. N. Couch, U. N. C.

x*Detection of fungus spores in the environment by means of the membrane filter. Occurrence of* Histoplasma capsulatum *and* Microsporum gypseum *in soil.* M. A. Gordon, USPHS, Atlanta, Ga.

Soil moisture and chemical composition of flue-cured tobacco leaves. C. H. M. van Bavel, N. C. State

x*The correlation of action potentials from Venus' Fly-trap with* Nitella. Otto Stuhlman, Jr., U. N. C.

x*Relation of light to the germination of dormant seed of flue-cured tobacco.* T. E. Smith, McNair's Yield Tested Seed Co.

x*Rapid growth of* Metasequoia. H. R. Totten, U. N. C.

x*Radiographic study of flowers.* E. Jane Cox, Woman's College U. N. C.

x*Effects of compounds with chemical similarities to maleic hydrazide on the growth of plants.* V. A. Greulach, U. N. C.

Translocation of P^{32} *from various regions of barley roots.* H. H. Wiebe and P. J. Kramer, Duke

x*A study of the cut-twig method for measuring transpiration.* C. J. Lilly, Mitchell College

Phytogeographical derivation of shrub-bog and swamp forest species. B. W. Wells, N. C. State

A new heart-leaf from North Carolina. H. L. Blomquist, Duke

A native habitat for certain urban mosses. L. E. Anderson, Duke

GEOLOGY SECTION

x*Geology of the Irmo, South Carolina, Quadrangle.* S. D. Heron, Jr., Duke

x*Recent changes in the shore line near Point Barrow, Alaska.* G. R. MacCarthy, U. N. C.

x*Solution or not.* E. W. Berry, Duke

Geology and industrial developments in North Carolina since 1940. J. L. Stuckey, N. C. State

MATHEMATICS SECTION

The deflection curve for beams with variable cross sections. T. F. Hickerson, U. N. C. (Published in full in this issue of the Journal)

The application of Fourier Series to the problem of Dido. Frances Wolfe, Woman's College, U. N. C.

On the characteristic roots of some matrices which are of interest in economics. Alfred Brauer, U. N. C.

Empirical equations illustrating inverse variation in the social sciences. Whitfield Cobb, Guilford College

Utilization of environment in the teaching of plane geometry. J. U. Crum, Western Carolina Teachers College

PSYCHOLOGY SECTION

(N. C. Psychological Association)

Review of Research

Woman's College U. N. C. Kenneth McCutchan
North Carolina State College. D. J. Moffie
Queens College. Ethel M. Abernethy
Duke University. E. H. Rodnick
University of North Carolina J. F. Dashiell

Presidential Address. Dorothy C. Adkins

ZOOLOGY SECTION

x*Records of fairy shrimps in North Carolina.* Tom Daggy, Davidson

x*Photoperiodism in the freshwater shrimp,* Palaemonetes paludosus (*Gibbes*). O. H. Paris and C. E. Jenner, U. N. C.

x*Investigations on the life cycle of the Tennessee brook lamprey.* E. C. Horn and J. R. Bailey, Duke

The making of keys. Z. P. Metcalf, N. C. State

Transcontinental observations of wildlife ecology. H. C. Jones, East Carolina College

Histology of digestive and excretory systems of Balanus amphitrite. C. G. Bookhout, Duke

Problems concerning the origin of vertebrates. T. H. Eaton, East Carolina College

x*Some protein changes during amphibian embryogeny.* G. W. Nace, Duke

x*An analysis of regulation in the amphibian neural crest.* H. E. Lehman and Lillian M. Youngs, U. N. C.

x*The role of ectoderm and mesoderm in amphibian pigmentation.* H. E. Lehman, U. N. C.

x*An investigation of maternal influence on the fused gene through ovarian transplantation between certain strains of the house mouse.* Robert Tamis and E. C. Horn

Segregation of harelip and the Laurence-Moon-Biedl syndrome in a single sibship. Maurice Whittinghill and Evelyn E. Hendricks, U. N. C.

x*The relationship of the comparative hematology of certain species of salamanders to their ecology.* F. J. Vernberg, Duke

Fecundity of Hudson River shad. B. A. Lehman, U. S. Fishery Laboratory

Some mimicry patterns in the pepsine spider wasps. Henry Townes, N. C. State

x*Eggs and young of the lizard,* Cnemidophorus sexlineatus (*Linnaeus*). E. E. Brown, Davidson

Abstracts:

Biochemical Control of Caste in the Social Life of an Insect Community. A. L. PICKENS.

During long eons of cryptobiotic and beleagured existence the termites have met and overcome many biochemical problems. *Reticulitermes,* our most common

genus, though attacked by *Termitaria pacedensis* and its relatives, controls a number of other threatening fungi and perhaps uses some as food. A queen, sterile on mere cellulose, may soon begin to lay when fed fungus infested wood. Long before man, these insects were evidently using yeasts to offset deficiency diets.

The term *caste* has been abused. From 14 to 16 sizes and forms may be found in *Reticulitermes*. Only three: reproductives, workers and soldiers are castes, that is, forms with distinctive shares in the group activities. If arranged like red, blue, and yellow on a color-wheel we will find three intercastes occupying the spaces of the secondary colors.

R

T N

E

S W

G

R = winged, or primary, reproductives; N = Neoteinies, or supplemental, reproductives; W = workers; G = "guerrillas" or the worker soldier intercaste; S = soldiers; T = tyros, or the soldier reproductive; E = egg mass from which all develop.

The reproductive-worker class is very important and supplies secondary egg-layers more efficient than the primary queen. They connect the last with the workers which are also potential reproductives. Neither secondary nor worker can lay eggs until freed of the inhibition of the queen who, through a secretion from her body, holds them in sexual immaturity. Upon her death, or when separated, the least inhibited quickly attain the reproductive condition and so inhibit the others. The other intercastes are worker-soldiers and soldier-reproductives. Soldiers appear to inhibit also, and thus an older soldier in a small colony would hold back from the soldierhood members hatching later, but one almost as old might attain a partial condition and be arrested in mid-career as half soldier, half worker. Much older groups in times of stress, when separated from the queen, may cannibalize their secondaries, and permit the last of the soldiers to perish. A group of workers left with neither inhibitive present may thus be found growing both toward soldierhood and reproductive condition at one time, so producing the rare "winged soldier" or soldier-reproductive.

Midget, dwarf, regular and giant soldiers and workers represent mere size variations in smaller or larger colonies with lesser or greater facilities for intercommunal feeding. In larger colonies they grow longer and reach additional instars for the terminal moult. It is worthy of note that in early swarms from young colonies two sizes of flying forms may emerge, resembling two species. Two sizes may be expected in one or more of the intercastes.

The inhibiting secretion has been extracted and fed in cellulose by associated

investigators who obtained favorable results with subject termites in reproductive tests. This secretion, current indications associate with the corpora allata or a similar organ.

An Optical Analysis of the Laminated Lens of a Typical Teleostat Eye. OTTO STUHLMAN, JR.

The refraction of the fish eye was treated in great detail by Matthiessen as early as 1879 (Pflüger Arch. Physiol. **19:** 480). It was found that the fish eye was more thoroughly standardized than any other. He showed that the refractive index of the spherical lens decreased with the square of the radius, and that the distance from lens-center to retina was 2.46 times the radius of the lens and independent of the size of the fish. The lens never changes its shape during accommodation. It can, however, be drawn backward by the retractor lentis to focus an image on the retina. In order to function at such a short focal distance, the lens must of necessity possess a higher effective index than the maximum index of refraction of its nucleus. From his limited data Matthiessen calculated the value of the nuclear index as 1.5215.

Using his observed mean value for the index of refraction of the cortical layer as 1.3830 and the law of decrease in index as the square of the radius, it can be shown that the focal length of the teleostat eye is too long for effective vision, since the image would fall outside the retina. No focusing mechanism is available to correct this farsighted condition, so the fish would be blind. To meet this theoretical impossible situation it was proposed to re-examine the available data.

The ideal ratio of focal length to radius, 2.46, was retained. The decrease in index of refraction with the square of the radius was accepted as basically correct for the variation of the index of refraction of the laminations of the spherical lens. The data for cod eyes were accepted as typical and the extrapolated value for the nuclear index was found to be 1.530 instead of 1.5215 as calculated by Matthiessen.

With the anterior surface of the eye emersed in water, index 1.333, and its posterior surface in vitreous of index 1.335, it can be shown that in such an ideal eye the ratio of focal length to radius is very close to 2.46, and is independent of the number of laminations, if greater than four. These conclusions were arrived at with the aid of the Gaussian forms of the single lens equation, using first order solution for paraxial rays.

Since the layers of circumferential lens fibers are laid down over the nucleons during growth and since focal length over radius does not vary with age, one may conclude that ageing of the lens fibers produces the necessary increase in index of refraction from 1.383, the original value of the new fibers, to the higher values found for the fibers surrounding the nucleons, thus constantly retaining the decrease in index proportional to the square of the radius of the spherical lens, and maintaining the ratio of focal length to radius of the lens as 2.46. The unaccommodated teleostat eye is therefore normal if the nuclear index of the lens is 1.530 or greater.

Serological Studies of the Blood of the Developing Chick Embryo. G. W. NACE.

The time of appearance of albumin, $\alpha\beta$- and γ-globulin in chick embryo serum was studied by means of interfacial precipitin reactions. Rabbit antisera to total adult chicken serum, albumin and γ-globulin, the latter prepared by cold ethanol fractionation, were absorbed with the heterologous serum fractions and/or with yolk, and appropriate specificity tests were made with these fractions and with yolk and its fractions, livetin and vitellin. These tests suggested that livetin contains components representing each adult fraction and that each adult fraction contains yolk-like constituents.

Chick blood and serum from the third day of incubation to the first day post hatching were tested with these antisera and the following description of embryonic serum differentiation was proposed: a) Embryonic serum antigens may be characterized as either vitelloid, i.e. yolk-like, or non-vitelloid, i.e. distinguishable from yolk. b) A vitelloid albumin is present from the third day onward. c) A non-vitelloid albumin-like material(s) resembling adult albumin may be present on the fifth day. d) A non-vitelloid albumin-like material(s) distinct from adult albumin seems to be present on, at least, the third and fourth days. e) A non-vitelloid $\alpha\beta$-globulin-like material(s) is detected on the sixth day. f) A vitelloid γ-globulin-like material(s) is present by the ninth day. g) A non-vitelloid γ-globulin-like material(s) arises about the twelfth day.

Evidence from the literature was correlated with these results and the possible significance of these constituents was indicated.

(Investigations carried out as a Public Health Research Fellow of the National Institute of Health.)

Fluoride and Total Liver Lipid. C. L. YARBRO, BRADFORD MILLER and C. E. ANDERSON.

In view of the present emphasis upon the fluoridization of public water supplies, further knowledge of the effect of fluoride upon intermediary metabolism seems important. This subject has been reviewed by Dori (Arch. f. Mineral o. Geol. 20A, No 8, 1948).

Fluoride (one part per thousand and upward) interferes with carbohydrate and fat metabolism. Hence it appeared reasonable to expect the appearance of fatty livers in animals receiving fluoride in comparatively high concentrations. Vellu and Zottner reported fatty livers in sheep which had died from drinking water with a high fluoride content. This finding was not confirmed by Pierce or independently by Phillips and Lamb using sheep and rats respectively.

Because of the conflicting data currently in the literature the following experiment was planned and carried out. Ten, thirty day old hamsters were fed a stock diet and water containing fluoride in a concentration of fifty parts per million. Five animals received distilled water in addition to the stock diet and served as controls. At the end of the four month experimental period the liver of each animal was analyzed for total lipid. No increase in total lipid was found in the fluoride treated animals when compared with the controls. This finding does not rule out an upset in the ratios of the various components of the liver lipids. Fractionation of the liver lipid is now in progress.

The Influence of Fluoride on Whole Body Retention of Radioactive Rubidium. M. K. BERKUT and J. C. ANDREWS.

Drea in 1936, and Lowater and Murray in 1937, following their spectrographic analyses of human teeth, reported incidental observations and spectrographic data, respectively, which suggested that the composition of enamel of human teeth originating in a high-fluoride area was different in its potassium content from enamel of teeth from animals that were never exposed to a fluoride environment. Our studies have been designed to observe, indirectly, the influence of fluoride administered as sodium fluoride on the mobilization or incorporation of potassium in calcifying incisor teeth of hamsters. Because of its longer half-life we have employed the use of radioactive rubidium (Rb^{86}), in multiple intraperitoneal injections, as a substitute for potassium. Work has recently been extended to studies of the effect of fluoride on the rate of excretion and whole body retention of radioactive rubidium in the hamster. Fluoride injections, subcutaneously and intraperitoneally, were given 20 hours after a single dose of isotonic rubidium carbonate containing induced radioactive rubidium had been injected. Hamsters which received the fluoride intraperitoneally averaged a total urinary and fecal excretion of only 5.3% of the total injected radioactivity, while controls that had received isotonic sodium chloride excreted 20.1% for the same period. Total urinary and fecal excretions as low as 0.01% were observed in some treatment animals, while controls for the same period excreted 5.8%. A mean value of 35.3% of the injected dose of radioactivity was retained in the carcasses of the fluoride injected animals as compared with 20.1% for the saline treated controls. At the level injected, no change in rate of rubidium excretion, or of rubidium retention, was provoked by subcutaneous fluoride. This was probably due to failure of the absorption of fluoride from the site of injection since severe edematous areas developed. No effect on the rate of excretion of radioactive rubidium was observed in animals that received 5 and 50 parts per million fluoride as sodium fluoride in their drinking water during the experimental period of 10 days.

(Work performed under Contract AT-(40-1)-299 with the U. S. Atomic Energy Commission)

Blood-Clotting Studies on Dogs Injected with Radio-Gold. J. H. FERGUSON.

When blood is removed from the body and clots, a plasma protein called prothrombin is converted into thrombin which then changes another plasma protein, fibrinogen, into the jelly-like fibrin, which is the essential substance of the blood clot. Normally, the conversion of prothrombin to thrombin continues after the clotting, so that practically all the prothrombin is used up ("consumed") within half-an-hour after clotting. If more than ten per cent of the prothrombin still remains in the serum 1–1½ hours after clotting, there is something defective about the "prothrombin consumption". This defect has been demonstrated (by Dr. E. P. Cronkite and colleagues) to occur in radiation injury following experimental atomic bombings and in animals exposed to fatal doses of X-ray with the 2-million volt X-ray machine at the U. S. Naval Research Laboratories. Dr. J. H. Ferguson of Chapel Hill has developed a new approach to these radia-

tion problems, in co-operation with researchers at the Oak Ridge Institute of Nuclear Studies. These workers employ radio-isotopes to try and locate the radiation damage to selected regions of the body, in an effort to obtain more exact data as to the particular factors involved in these blood clotting abnormalities. Radio-gold was selected for the first of these studies because it is deposited in the liver and spleen almost exclusively. The liver is known to form several of the proteins which are important for blood coagulation. About the sixth day after the injection of radio-gold, in sufficient dosage, the prothrombin consumption test becomes abnormal, although no change may be detected in simpler clotting tests. The test results get worse until the animal dies, or improve, after a period, if the dog recovers. Preliminary results suggest that the new test may be of practical value in the early detection of severe radiation injury and may be sufficiently quantitative to follow the course and perhaps predict the outcome. Thus, the test may be a valuable tool when used on human cancer patients undergoing radiation therapy. The experiments on dogs receiving radio-gold injections are being reported at the April meetings of the Federation of American Societies for Experimental Biology, New York, 18 April, 1952.

Further Studies on the Motile Spores of Actinoplanes. W. J. KOCH and J. N. COUCH.

It has previously been reported by Couch that the planospores of *Actinoplanes* are more like motile bacteria than fungus zoospores. No flagella are apparent with ordinary bright field or phase microscopy. The spores swim very rapidly and when examined in a dark field have the appearance of short coils with about five complete turns.

Such spores have been photographed at shutter speeds of 1 second to 1/100 of a second. These photographs indicate that the coiled appearance of the rapidly swimming spore results from its helical path. There are two components of spiral movement. The spore describes a greater spiral as it moves in its tight helical path. The speed of fast-swimming spores has been calculated.

The planospores show an apically attached tuft of bacterial-like flagella after staining by bacteriological methods or when observed with the electron microscope. There is evidence of interspecific differences in the average size and internal structure of the planospore.

Detection of Fungus Spores in the Environment by Means of the Membrane Filter. Occurrence of Histoplasma capsulatum *and* Microsporum gypseum *in Soil.* M. A. GORDON.

The membrane filter affords a simple, rapid, and efficient method for the detection of fungus spores in hydrosols and aerosols. The exposed membrane may be subjected to a selective stain and examined through the compound microscope by means of transmitted light. Details of this procedure are described.

Whatman filter papers of certain grades and Seitz filters may serve as acceptable substitutes for the more expensive and less generally available membranes and special membrane filter apparatus.

In field trials of this method, typical tuberculate spores of *Histoplasma capsulatum* have been recovered from several soil samples and from river water. Macroconidia of *Microsporum gypseum* have also been found in soil.

A method is described for preparing stained permanent transparent mounts of the filter discs.

The Correlation of Action Potentials from Venus' Fly-Trap with Nitella. OTTO STUHLMAN, JR.

From an analysis of the action current obtained from a single cell of *Nitella*, Hill and Osterhout (Jour. Gen. Physiol. **18:** 215, and 377, 1934/35) concluded that the general features of the observed action potential were attributable to the movement of potassium ions accompanied by an increase in permeability. Their action potential shows an initial rapid rise and fall from a sharp maximum potential, followed by a similar though broader maximum, which dropped slowly with a speed which led them to conclude that the recovery was primarily a process of diffusion.

The action potential obtainable from Venus' fly-trap is similar in structure, and can be resolved into two components. These two components can be shown to vary independently, and to possess different speeds of propagation across the leaf of the plant. The activity seems to indicate the existence of a breakdown of a polarization, independent of the potential changes accompanying the movement of an ionized fluid. The latter moves across the leaf more slowly than the former. The relative displacement of the two phases was found to be controllable, varying with the position of the electrical contacts on the surface of the leaf. The depolarization action can be excited to lag or lead the hydrodynamic activity. The latter could even be suppressed, leaving only depolarization action potentials as found in simple mammalian nerve activity.

Relation of Light to the Germination of Dormant Seed of Flue-Cured Tobacco. T. E. SMITH.

Certain lots of tobacco seed have been observed with low germination within a few weeks after maturity and high germination after storage. Seed grown in 1951 provided material to study this problem. In the germination tests seed were spread on moistened blotters in petri dishes and incubated at room temperature usually for ten days. Seed of Dixie Bright 102 germinated to the extent of 6% in darkness, 62% in diffuse light in the laboratory, and 93% in three hours of sunlight each day. Seed of the 402 variety germinated 0.0% in darkness and 97% on a white blotter in diffuse light of the laboratory. Further tests showed that the light requirement for germination occurred in new crop seed much more often than in seed one or more years old.

Rapid Growth of Metasequoia. H. R. TOTTEN.

The genus *Metasequoia* was described and named by the Japanese botanist Miki in 1941, who based his description on fossil material from Korea and Japan. The genus was thought to have been extinct for about twenty million years.

However, only about four years later, in 1944 or 1945, specimens were collected from living plants in China by Mr. Tsang Wang, of the Central Bureau of Forest Research in China. Other Chinese botanists, among them Dr. H. H. Hu, examined Wang's specimens and recognized them as belonging to the recently named genus *Metasequoia*. Dr. Hu described the species, giving it the name *Metasequoia glyptostroboides*. Botanical specimens reached Dr. E. D. Merrill of the Arnold Arboretum late in 1946. He secured a small grant ($250.00, but equal to $9,750,000 in Chinese currency) and sent it to Dr. Hu for an expedition to secure seeds. This expedition was sent out by Dr. Hu on September 3, 1947, and the seeds arrived at the Arnold Arboretum on January 5, 1948. Dr. Merrill and the Arboretum sent seeds to many arboreta in this county and abroad, some of them to the Coker Arboretum in Chapel Hill. Since that time quite a number of papers have appeared on *Metasequoia*, most of them mentioning the rapid growth of the plant.

Dr. Donald Wyman, of the Arnold Arboretum, in the November, 1951, number of *Arnoldia*, has a picture of a 28-month-old plant grown in a coldhouse at Bramley, England, that was 8 feet 3 inches high; and a picture of one in the Arnold Arboretum from seed sown in July, 1948. The plant, when transplanted out of doors in the spring of 1951, was 4 feet, 4 inches tall, and when the photograph was taken, the plant which was slightly over three years old, measured 8 feet, 8 inches in height.

At Chapel Hill, from 30 seeds planted in the spring of 1948 we secured one seedling. That summer it was transferred to a flower pot and it wintered in the greenhouse. The following spring it was planted outdoors. At the end of the 1950 season it was 8 feet, 7 inches high; and at the end of the 1951 season 12 feet, 9 inches high. In March of this year Mr. Howard W. Blakeslee in widely published newspaper articles credits us with leading the race in the rapid growth of *Metasequoia*, giving second place to a plant in Santa Barbara, California, ours leading by six inches. However, Dr. R. W. Chaney, Professor of Paleontology of the University of California, who has worked considerably with both the fossils and the living plants of the genus, and who has also collected it in the Szechuan and Hupéh Provinces of China, will shortly publish, if he has not already done so, a report of a plant considerably higher than ours growing at Pasadena, California. Volume **34** of the *Proceedings of the National Academy of Sciences*, November, 1948, carries a very interesting paper by him, entitled "The Bearing of the Living *Metasequoia* on Problems of Tertiary Paleobotany."

Radiographic Study of Flowers. E. JANE COX.

A brief explanation was given of x-ray physics with reference to the necessary wave lengths of the electro-magnetic spectrum and the inherent filtration of these wave lengths. The internal structure and beauty of botanical subjects was demonstrated with slides, x-ray negatives, and positive paper prints.

Effects of Compounds with Chemical Similarities to Maleic Hydrazide on the Growth of Plants. VICTOR A. GREULACH.

Except for maleic hydrazide, the hydrazides, imides and similar compounds have not been investigated for their effects on plant growth. The few studies of the effects of maleic acid on plants are somewhat contradictory, indicating that it may retard ripening of apples and pears and sprouting of potatoes, inhibit plant growth slightly, and inhibit the activity of proteases in fig latex. On the other hand, it has been reported to be utilized by wheat roots and to counteract the effect of iodoacetate on *Avena*. Somewhat more extensive work with maleic acid on animal tissues suggests in general that maleic acid inhibits respiration and growth, though there are also reports of the use of maleic acid by animals and animal tissue. Since maleic hydrazide is an antimitotic, it is interesting that Friedman *et al.* (*Brit. Jour. Pharmacol.* **3:** 263, 1948) found maleic acid to be an antimitotic in chick fibroblasts and later (*Idem* **4:** 105, 1949) reported that maleimide and other unsaturated imides were even more effective, though succinimide was not. They claimed that the antimitotic effectiveness of the compounds depended on their—SH uptake.

Tomato, sunflower and bean plants were treated by dipping young plants in solutions of maleic hydrazide, succinic hydrazide, diformyl hydrazine, succinimide, phenylhydrazine hydrochloride, isonicotinic hydrazide (rimifon), 1-isonicotinyl 2-isopropyl hydrazide (marsilid), sodium maleate, sodium succinate, and sodium fumarate. Except that 0.03 diformyl hydrazine was used in one experiment with beans and that the quite insoluble succinic hydrazide was applied as a saturated solution (about 3×10^{-4} molar), all solutions were 0.015 molar. Some bean and tomato plants were also treated with 4000 p.p.m. succinic hydrazide in lanolin, applied to the under side of one leaf.

Except for phenylhydrazine hydrochloride, none of the substances produced growth inhibition as marked as that caused by maleic hydrazide. The phenylhydrazine killed the younger tissues, but most plants later resumed growth, although the new leaves were about half the usual size. While 0.015 M diformyl hydrazine did not inhibit growth, 0.03 M did inhibit the growth of beans. The upper surface of the bean (but not tomato) leaves dipped in both concentrations of diformyl hydrazine turned white owing to the loosening of the epidermis. Succinic hydrazide failed to inhibit tomato plants, but did inhibit the growth of bean plants, which were spindly and had general chlorosis, but lacked all of the maleic hydrazide symptoms. Both rimifon and marsilid inhibited the growth of all three species, though sunflower plants recovered rapidly from rimifon inhibition. Marsilid was generally more effective than rimifon, both in inhibiting growth and in producing formative effects, and caused loss of apical dominance and branching in about a third of the sunflower plants. Formative effects in sunflowers included downward or upward curling of leaf margins, emarginate and wrinkled leaves, and very narrow and distorted young leaves. Similar effects were produced in bean plants, but there were no formative effects in tomatoes. All plants treated with succinimide, succinate, maleate, and fumarate were more vigorous than the controls and in one experiment with beans the succinimide and maleate produced a significant increase in growth over the controls. All growth inhibition and promotion mentioned was significant at the 1% level. Additional work with these and other related compounds is in progress.

A Study of the Cut-Twig Method for Measuring Transpiration. C. J. LILLY.

The cut-twig method for measuring transpiration was evaluated by comparison with the conventional gravimetric procedure. The water loss of a potted plant was determined for 30 to 60 minutes on a triple beam balance, then the stem was cut and the rate of water loss of the excised shoot was determined for 10 to 21 minutes with an analytical balance. Species used in the 44 tests were white oak (*Quercus alba* L.), black oak (*Quercus velutina* Lam.), chestnut oak (*Quercus prinus* L.), northern red oak (*Quercus rubra* L.), green ash (*Fraxinus pennsylvanica* var. *subintegerrima* (Vahl) Fern.), short-leaf pine (*Pinus echinata* Mill.), tulip poplar (*Liriodendron tulipifera* L.), and geranium (*Pelargonium zonale* Willd.). In addition, 20 transpiration tests were made with cut twigs from large trees of white oak, southern red oak (*Quercus falcata* Michx.), tulip poplar, sugar maple (*Acer saccharum* Marsh.), and American elm (*Ulmus americana* L.).

Transpiration rates for excised shoots usually were less than those for the corresponding intact plants. Average rates for excised shoots by species always fell below those of the intact plants. Transpiration rates for the cut twigs generally declined with time after cutting, but some exceptions to this were noted. Constant rates of water loss for a period of three minutes after the initial weighing of the excised shoot were exceptional. Determinations with cut twigs from large trees support the results obtained with excised shoots of potted plants. Transpiration rates of excised shoots were averaged by species for one-, three-, and ten-minute periods. High standard deviations indicate much variability in the replicate tests used in obtaining average transpiration rates, and indicate that the species differences in transpiration rates for excised shoots have doubtful significance.

The results of these tests strongly suggest that for the native woody species investigated the cut-twig method for determining water loss is not a reliable index to the rate of transpiration prior to excision.

Geology of the Irmo, South Carolina, Quadrangle. S. D. HERON, JR.

The Irmo Quadrangle is located in the Piedmont and Coastal Plain physiographic provinces of central South Carolina. The Carolina Slate is the principle formation of the Quadrangle. It has been intruded by four separate areas of granite, two quartz-diorite dikes, a hornblendite, and numerous diabase dikes. The Tuscaloosa Formation overlaps part of the southern portion of the Slate, and alluvium covers the valley floors. Metamorphosed volcanic tuff and breccias make up the bulk of the Carolina Slate, but near the Spillway Granite, it has been altered to a coarse garnet mica schist. Other intrusive contacts are hidden because of the deep and thorough weathering. The granite bodies of the area are, for the most part, small and coarse-grained. Two show a slight lineation in both the hand specimen and the thin section. The Spillway Granite has a definite gneissic appearance which is primary in origin. The fourth and largest granite has the composition of a granodiorite, though little structural detail can be obtained from this granite because of the deep weathering. The Tuscaloosa Formation consists of sand and sand-clay mixtures of Upper Cretaceous age.

Recent Changes in the Shore Line near Point Barrow, Alaska. G. R. MACCARTHY.

Most of the shore lines in the neighborhood of Point Barrow, Alaska, exhibit marked evidence of retreat. This evidence consists in part of numerous small hanging gulleys in the sea cliffs, fresh slumping and falls from the face of the bluffs, numerous transected lake basins, and the destruction of several triangulation stations established by the U. S. Coast and Geodetic Survey as recently as 1945. At several places it has been possible to measure the exact amount of change which took place between 1945 and 1949, and between 1949 and 1951. Coastal erosion in this area is greatly influenced by the presence of ground ice, particularly by the vertical ice wedges associated with the development of polygonal ground.

Solution or Not. E. W. BERRY.

The Trent and Castle Hayne Marls of eastern North Carolina have a most irregular upper surface due to solution cavities on the top of the marl. In places, however, there are vertical "pipes" of sand and clay which seldom reach more than a foot in diameter, and appear to extend to a considerable depth in the formation; in most cases the bottom is not exposed. These restricted, long vertical "pipes" are due to channeling by upward rising water and/or gases during the compacting of the original marls, thus localizing a place where subsequent solution could penetrate entirely through the marls. After compaction, solution widened these holes slightly and they became filled with less calcareous materials.

Records of Fairy Shrimps in North Carolina. TOM DAGGY.

No records of fairy shrimps (Crustaceans of the Order Anostraca) in North Carolina have previously been published. In 1951 and again in 1952, two species, *Eubranchipus vernalis* (Verrill) and *E. holmani* (Ryder), were collected in vernal pools on the flood plain of Rocky River in Cabarrus County, North Carolina. Published records place *E. vernalis* from Ontario to British Columbia and south to Illinois and New Jersey. In a personal communication H. C. Yeatman reported collecting this species near Chapel Hill, N. C. *E. holmani* has been recorded from Massachusetts to Ohio and south to Virginia and Tennessee, with two disjunct records from Louisiana. In a personal communication Pearse reported that Gray and Bookhout had collected an undetermined fairy shrimp in Duke Forest near Durham, N. C.

The following data apply to the Cabarrus County collections. *E. vernalis* was taken from two adjacent pools. In the first pool it was abundant (1,218 specimens taken in all), the specimens were bright in color and averaged 16.2 mm in length. In the second pool it was scarce (16 specimens), darker in color, and averaged 21.2 mm in length. *E. holmani* was taken only from the first pool, a total of 23 males being collected (the females were not separated from *E. vernalis*). Sampling in a number of other likely pools in both Cabarrus and Mecklenburg Counties failed to produce fairy shrimps.

Photoperiodism in the Fresh-Water Shrimp, Palaemonetes paludosus (Gibbes). OSCAR H. PARIS and CHARLES E. JENNER.

In a preliminary study of photoperiodism in the fresh-water shrimp *Palaemonetes paludosus,* one group of 17 shrimp (sex undetermined) was placed on a long day (18 hours of light, 6 hours of dark), and an equal number was placed on a short day (9 hours of light, 15 hours of dark). Animals were cultured in light-proof chambers provided with "daylight" fluorescent lamps (intensity approximately 30 f. c.). The results of the experiment, which was continued for three months, are shown as follows:

Short-day shrimp: 1 female underwent ovarian development; 8 females underwent no ovarian development; 5 shrimp died (sex undetermined); 4 shrimp died after end of experiment (sex undetermined).

Long-day shrimp: 11 females underwent ovarian development; 3 females underwent no ovarian development; 3 shrimp died (sex undetermined).

A second experiment was designed to confirm the results of the first and also to test the importance of the dark period as the critically important factor. Three groups of shrimp consisting of 10 females and 4 males were placed under each of the following conditions: a long day (18 hours of light, 6 hours of dark), a short day (11¼ hours of light, 12¾ hours of dark), and a short day with the dark period interrupted with a brief period of light (9½ hours of light, 6½ hours of dark, 1½ hours of light, 6½ hours of dark; a total of 11 hours of light per 24 hours). Within 72 days, all female survivors (8) on the long day and all 10 females on the dark-period interruption had undergone ovarian development. At the end of the experiment (90 days from start) no females on the short day (all survived) had shown any ovarian development.

These experiments represent the first time to the writers' knowledge that photoperiodism has been clearly demonstrated in the Crustacea. The demonstrated importance of the dark period supports the theory that dark-period dependence is characteristic of all photoperiodic responses, plant or animal.

Investigations on the Life Cycle of the Tennessee Brook Lamprey. EDWARD C. HORN and J. R. BAILEY.

A total of 555 ammocoetes of the non-parasitic Tennessee brook lamprey, *Ichthyomyzon hubbsi* Raney, have been taken during different seasons from the Mills River, Henderson County, North Carolina. Of these, four were metamorphosing specimens. A number of specimens from a January collection have been kept alive in the laboratory, but none has metamorphosed.

Graphing the size distribution of each collection according to the month taken has led to the following conclusions: (1) Size groups for the first and second year classes are clearly distinguishable, beyond which they become obscured by excessive overlap; (2) transformation of the ammocoetes may occur in the third year; (3) a period of little or no growth may precede transformation in some ammocoetes, extending the larval period to the fourth year; (4) adults of the species and our metamorphosing specimens average considerably less in length than the large ammocoetes, suggesting reduction in size during transformation; (5) transformation probably occurs in the fall of the year preceding spawning.

Some Protein Changes During Amphibian Embryogeny. G. W. NACE.

Serological and electrophoretic methods have been utilized to investigate the early development of *Rana temporaria*. Antisera to mature ovarian oocyte and hatched tailbud fractions and to adult frog sera were injected into one of the first two blastomeres and the blastocoel, and were introduced into neural crest cultures and into media in which embryos were developing. The only specific effects observed were with anti-tailbud supernatant. Gastrulae and neurulae cultured in dialyzed and diluted anti-tailbud supernatant showed an inhibition of development, gradient-wise cytolysis and death at stage 19–20. These results demonstrate an antigen which arises between the oocyte and tailbud stages and resides in the epidermis at stage 19–20.

Unabsorbed and appropriately absorbed antisera were used in interface precipitin reactions with test antigens from various stages of development. Both serum-like and non-serum-like combining groups were found throughout development; the vitellin fractions were serologically identical in the oocyte and tailbud stages; and two new antigens were detected, one in the mature ovarian oocyte supernatant, the other in the tailbud supernatant.

Electrophoresis of the supernatant fractions provides evidence for the existence of two major components throughout development, with the exception of the immature ovarian oocyte where one major component was observed.

The essential agreement of the biological, *in vitro* serological and electrophoretic experiments indicates the appearance of at least two new proteins during development.

(Investigations carried out as a Public Health Service Research Fellow of the National Institute of Health)

An Analysis of Regulation in the Amphibian Neural Crest. H. E. LEHMAN and L. M. YOUNGS.

The senior author recently reported (*Biol. Bull.* **100:** 127–151, 1951) an intrinsic difference in the pigmentary component of the neural crest in two species of Amphibians. This difference concerned the capacity of the pigment primordium to compensate for partial extirpation of crest material. In *Triturus torosus* the melanophore population was reduced approximately 40%, indicating a low degree of regulation; in *Triturus rivularis*, there was no difference in melanophore populations at late stages of development, indicating a high regulatory capacity. The present study is a continuation and an extension of the analysis of regulation in embryos deprived of half their normal complement of neural crest material.

Neural fold was extirpated unilaterally at open neural plate stages in two species of salamanders, *Triturus torosus* and *Amblystoma punctatum*. The resulting experimental embryos thus contained only one-half of their normal complement of neural crest material. Regulation of two crest derivatives, melanophores and dorsal root ganglion cells, was quantitatively determined by counting these cells during progressive stages in development, and comparing "half-crest" numbers with control numbers.

In *T. torosus*, the primary melanophore population of half-crest embryos was reduced approximately 50% at Twitty stage 37. By 15 mm. stages there was no

increase in primary melanophore numbers either in control or half-crest larvae, and the reduction of primary melanophores was still approximately 50%. As far as could be determined by these experiments, the neural crest of *T. torosus* is capable of contributing a very restricted number of primary melanophores. These cells are those which complete their differentiation between stages 34–41, and form the characteristic dorsal bands of the torosus larvae. Moreover, their quantitative determination seems to be quite rigidly established as early as the mid-neurula stage of development. The apparent rigid determination of primary melanophores, coupled with the relatively short period of their differentiation, is the means whereby the lack of regulation in these cells is expressed. In contrast, *A. punctatum* melanophores show an initial reduction of approximately 35% in half-crest embryos, but are capable of completely overcoming these deficiencies by late larval periods. Recovery of melanophore numbers of half-crest larvae is complete by 30 mm. stages; the production of melanophores is continuous.

Ganglion cells of *T. torosus* showed a reduction of approximately 35% in half-crest larvae at stage 43. Regulation in these derivatives was evidenced by the fact that at 18 mm. stages, the ganglion cells showed only a 20% reduction in the half-crest animals. The ganglion cell numbers of *A. punctatum* half-crest larvae showed an approximate 30% reduction at stage 44, while at 30 mm. stages the reduction was approximately 10%. Although the ganglion cells of *A. punctatum* are capable of regulation, this capacity proceeds at a slower rate than that evidenced in melanophores in these animals. Ganglion cells of *T. torosus* exhibit approximately the same regulatory capacity as ganglion cells of *A. punctatum*.

The Role of Ectoderm and Mesoderm in Amphibian Pigmentation. H. E. LEHMAN.

It has long been known that the number of pigment cells which differentiate in amphibian embryos is influenced by the ectoderm and mesoderm which provide the terrain in which these cells develop. Differences in pigment promoting capacity of ectoderm and mesoderm have been shown to vary from species to species, from age to age, and at different body levels. Previous studies have, almost without exception, dealt exclusively with the dark brown or black melanophores which differentiate during late embryonic and early larval stages.

The purpose of the present study is to examine the role of ectoderm and mesoderm in the development of other types of pigment cells which also differentiate in amphibians. The species used in the following experiments was *Amblystoma punctatum*.

The types of pigment cells which develop in this species are all derivatives of the neural crest and are distinguishable into four distinct groups: 1) dermal melanophores, melanated cells which begin to appear before feeding and, in general, are larger and more densely pigmented than the following group; 2) epidermal melanophores, melanated cells which appear first during early larval stages and are minute highly branched cells; 3) xanthophores, bright yellow cells which appear during pre-feeding stages; and 4) guanophores, silver cells which do not appear in any appreciable numbers until feeding stages. As for distribution throughout the body, dermal and epidermal melanophores and xantho-

phores are in the main restricted to the dorsolateral areas of the body, and are completely absent from the belly region during larval stages. Guanophores are widely distributed in peritoneal linings of the coelom and pericardium; however, during larval stages their only superficial distribution is under the epidermis of the lateral flank region, being absent from dorsolateral and mid-ventral regions.

The following experiments were carried out to discover whether or not the ectoderm and mesoderm play any part in bringing about this characteristic distribution of pigment cells. Over 400 grafts have been prepared with at least 10 embryos in each of the following 24 types of graft combinations. Ectoderm alone, and ectoderm and mesoderm, were taken from dorsal, lateral, and ventral locations on embryos at four different ages, namely, Harrison Stages 30, 33, 37, and 40. These tissues were individually grafted mid-dorsally onto young tailbud embryos. In order to exclude the possibility of transferring any prospective pigment cells with the grafts, the following precaution was taken in preparing donor embryos. The neural folds (pigment anlagen) were removed from prospective donors during open neural plate stages which rendered them neural crest-free and therefore pigment-less throughout their development.

The results of these experiments indicate that at mid-tailbud Stage 30 the entire ectoderm is essentially uniform in its capacity to promote the differentiation of dermal and epidermal melanophores and xanthophores; it is also uniform in its inability to foster guanophore differentiation. By late tailbud Stage 33 the ectoderm has become regionally different in its pigment promoting properties. From this stage on, dorsal ectoderm alone retains the chromogenic properties of earlier stages; whereas, lateral and belly ectoderm inhibit the differentiation of all types of pigment cells.

From the earliest stages tested (Stage 30) the mesoderm exhibits regional differentiation in pigment promoting properties. To a moderate degree all parts of the mesoderm at Stage 30 are capable of fostering the differentiation of dermal and epidermal melanophores, and xanthophores; however, only lateral mesoderm is capable of promoting the differentiation of guanophores. Following Stage 33, ventral and lateral mesoderm sharply inhibit the differentiation of dermal and epidermal melanophores and xanthophores. Even at older donor stages, guanophore differentiation occurs only in the presence of lateral mesoderm.

It is concluded that the normal distribution of pigment cells in *Amblystoma punctatum* is largely brought about by regional differences in the pigment promoting properties of the ectoderm and mesoderm, rather than being due to any intrinsic migratory differences in the individual types of pigment cells. For example, the distribution of dermal and epidermal melanophores and xanthophores appears to be controlled by regional differences in the ectoderm which become manifest in late tailbud stages. The distribution of guanophores during larval stages seems, on the other hand, to be controlled by regional differences in the mesoderm which are already present at the earliest period tested (mid-tailbud Stage 30).

These findings apply to the distribution of pigment cells during larval stages only. With the beginning of metamorphosis there are widespread changes in the

pigment pattern of the animal. Guanophores normally begin to appear dorsally in large numbers and after metamorphosis they contribute extensively to the "marbled" pattern characteristic of the adult. At this time also, melanophores and xanthophores invade lateral and belly regions. A similar sequence of pigmentary change is detected in the grafts during metamorphosis. These changes are in all respects topographically characteristic of the donor site of origin.

An Investigation of Maternal Influence on the Fused Gene through Ovarian Transplantation between Certain Strains of the House Mouse. ROBERT TAMIS and EDWARD C. HORN.

In an attempt to discover the possible site of maternal influence (hypothesized by Reed, 1937) affecting the expression of Fused, transplantation of Fused ovaries between the ovarian capsules of different strains of mice have been made. Strains were selected which would give a reasonably reliable check on the source of the offspring, i.e. whether from a donor or regenerated host ovary.

The resulting data are contradictory, suggesting the need for a revised hypothesis to explain the effect. Different ovarian transplantations are being made to test an hypothesis of the authors which depends on the mutual re-enforcement or counteraction of host and donor modifying factors.

Attempts have also been made to implant ovaries directly into the uterine horn. Thus far, no transplants have taken.

The Relationship of the Comparative Hematology of Certain Species of Salamanders to their Ecology. F. J. VERNBERG.

Red blood cell size of seven species of salamanders was measured during the spring months. Members of the same genus, as well as species, had cells of distinctly different sizes. In closely related species, the more active species had smaller red blood cells. Cold caused a change in the size and shape of the red blood cells of *Ambystoma maculatum* and *A. tigrinum*, but no increase in their number. Seasonal differences in size, shape, and number of red blood cells of *Plethodon cinereus cinereus* were observed. In general, an inverse relationship existed between number and size of red blood cells.

Blood sugar levels of six species of salamanders was determined. The range of the average levels was from 31.7–7.4 mg. per cent. A correlation between blood sugar level and activity was noted for these salamanders examined. The more active species had a higher blood sugar value than less aggressive forms. The level was markedly reduced in *Ambystoma maculatum* and *A. tigrinum* when these animals were subjected to cold.

Eggs and Young of the Lizard, Cnemidophorus sexlineatus (*Linnaeus*). E. E. BROWN.

Nests in considerable number are found in sawdust piles in the vicinity of Davidson. The average nest-cavity measures 2 by 2 in., with a depth of 1 in. It is located 4 to 5 in. (extremes, 3 to 11 in.) beneath the surface of the pile, within the zone of moist sawdust. An average set of 3 eggs (extremes of 55 sets, 1 to 5

eggs) occupies this cavity loosely and is not brooded by the female. 128 eggs carried through the incubation period in moist sawdust in the laboratory exhibited a mortality of 15%. The average ellipsoidal, whitish egg measures at laying about 16.5 by 9 mm. and weighs 0.85 gm. Just before hatching this egg measures about 18.5 by 13.5 mm. Among 45 sets of eggs the earliest date of hatching was June 27, the latest September 5. Eighty-two per cent of these sets hatched between July 10 and August 22. Present incomplete evidence suggests an incubation period very close to 8 weeks. 108 young from 38 broods included 56 males and 52 females. An average newly-hatched young lizard has a snout-vent length of about 32 mm., a total length of about 92 mm., and a weight of 0.84 gm. An average brood mother weighs about 9 gm. (without eggs). She lays a set of 3 eggs weighing 2.55 gm. (28% of her weight), and from her eggs hatches a brood of young weighing 2.52 gm. (28% of her weight).

REINARD HARKEMA, *Secretary*

PROCEEDINGS OF THE ELISHA MITCHELL SCIENTIFIC SOCIETY

1951–1952

490TH MEETING, OCTOBER 9, 1951

WHITTINGHILL, MAURICE (Zoology): *Experiments on Mendel's First Law.*[1]

Mendel's first law implies that a hybrid will hand on impartially any two genes of a heterozygous pair unless selection intervenes after gametes have been formed. Before gamete formation selection may be possible according to one of the hypotheses emerging from the author's study of crossing over as induced in *Drosophila melanogaster* by irradiation and by chemicals. On the assumption that induced crossing over would make sectors in the gonads like "twin spots," some cells being homozygous for normal genes and other cells being homozygous for lethal genes, extensive studies were made involving well over 100,000 testcross offspring. Isogenic wild (normal) stocks were prepared and then mated to a lethal-bearing stock, either Curly Lobe[4] or else Curly Pufdi. F_1 adults were treated with X-ray doses of 1500 r, 2000 r, 3000 r or 4000 r and then mated individually to light-eyed males in a sequence of cultures. Some untreated females, and also male heterozygotes, gave control data on the viability of the lethal-bearing chromosome as compared with the normal wild type during development of adults. The inheritance of the known lethals tended to decrease as the frequency of induced crossovers increased, reaching peaks in cultures three (from egg-laying 7–9 days after x-raying) and approximating those peaks in cultures four (9–11 days after treatment). Both values converged toward normal in the fifth cultures. The changes were more pronounced at lower doses, and in two instances were better than expected. This indicated that processes other than or additional to homozygosis in oögonial cells were operating. Whereas controls had 48.1 ± 0.44% of their non-crossover offspring belonging to the lethal-bearing class, females treated by 1500 r or 2000 r produced only 42.8 ± 1.26% and 43.0 ± 0.75% of Curly light Lobe (or Pufdi) from cultures three (7th–9th days). Experiments of improved design are now being started using more lethals in the one chromosome and extending the range of irradiation dosages.

H. D. CROCKFORD (Chemistry): *Standard Potentials in Aqueous Hydrochloric Acid Solutions Containing Organic Compounds.*

All electromotive force data for cells of the type:

$$H_2 \mid HCl(m), S(X), H_2O(Y) \mid AgCl\text{-}Ag$$

have been studied for the purpose of determining possible correlations between the standard cell potentials and the dielectric constants of the various solvent

[1] This study was aided by a research contract with the Atomic Energy Commission. The assistance of Alice Ryland Giles, Norma Sue Sligh and Margaret McBride Stewart under this contract is gratefully acknowledged.

solutions. The organic compounds, S, considered were d-glucose, d-fructose, dioxane, methanol, ethanol, propanol-2, glycerine, and ethylene glycol. Plots of E^0_m versus $1/D$ and (E^0_N–0.0591 log N_1) versus $1/D$ show definite similarities for similar types of compounds although the order of position is different in the two sets of curves.

Detailed studies have been made employing the various plotting procedures that can be applied to the potentials of cells of the above type for the purpose of determining the standard cell potentials. From these studies certain general conclusions can be drawn concerning the values of the ion size parameter and the "salting-out" constant for the various solutions. Some similarities may be drawn concerning these magnitudes and the positions of the curves in the plots referred to above.

491ST MEETING, NOVEMBER 13, 1951

JOHN F. DASHIELL (Psychology): *A Case of Fatigue-Induced Monocular Diplopia.*

A case is reported in which the visual images of letters and other objects appear doubled in the left eye's field and doubled then tripled in the right. This observation is made not with rested or normal vision but after protracted reading. Examinations of the literature of ophthalmology and of optics have unearthed no similar cases where multiple monocular imagery follows only after eye-work. Inquiries of authorities in those fields are likewise unrewarding. Yet inquiries among laymen bring to light corroborative cases.

Is the explanation to be sought in optical mechanism of eyeball, interactions or 'field' processes of the retinal elements, of the optic nerve fibers, of the lateral geniculates, or of the cortical elements? The first-named seems the most promising. The known facts about irregularities of lens shape (lenticonus, etc.), about pinhole vision, about the role of the iris in distinct vision, invite attention to the multiple cones of light cast by imperfectly symmetrical lenses which produce indistinct or blurred vision.

A theory advanced to explain the fact that the multiple imagery appears only after 'fatigue' is (1) that where lens irregularities interfere with clear vision the ciliary process reflexly makes some corrective adjustments involving radiating muscle fibers and (2) that prolonged work would lead to some 'fatigue' and relaxation of the corrective adjustments, and so to the production of multiple images not 'normally' present.

R. C. BOSE (Mathematical Statistics): *The Design of Experiments.*

492ND MEETING, DECEMBER 11, 1951

H. H. JAFFE (Experimental Medicine): *Theoretical Calculation of Hammett's Substituent and Reaction Constants.*

The effect of a substituent X on the reactivity of a side chain in the compound XC_6H_4Y is given by Hammett's equation. It was shown that it is possible to correlate the substituent constants entering in this equation (σ) with electron

densities calculated by the molecular orbital method in the LCAO approximation. For a series of compounds in which X contributes one π-orbital with two electrons (X = F, Cl, Br, I, OH, SH, NH_2) to the resonating system of the benzene ring, electron density contour diagrams were constructed in terms of Coulomb and resonance integrals as empirical parameters. By use of these contour diagrams a pair of parameters was assigned each substituent, and it was shown that these parameters were reasonable, and permitted calculation of mesomeric moments, absorption spectra and other physical and chemical properties in reasonable approximation. Substituent constants for toluene and naphthalene could also be calculated independently from the same assumptions, in good agreement with experimental values.

Introduction of a further assumption, which could be justified on the basis of experimental data, permitted calculation of relative values of the other parameter involved in Hammett's equation, the reaction constant (ρ). Actual calculations in a few selected cases showed good agreement with experimental results.

D. G. HUMM (Zoology): *Biochemistry of Growth in Salamanders.*

493RD MEETING, JANUARY 8, 1952

J. H. BROWN (Physiology): *The Metabolism of Methionine.*

The amino acid methionine has been widely used in the past few years in an attempt to treat various liver diseases and to improve nitrogen balance of individuals on a low caloric diet. In an attempt to explore the effects of various levels of methionine rats were fed the amino acid in a synthetic diet. At a level of 0.35% the animals had a higher nitrogen balance and gained faster than controls receiving the same diet but without the methionine. When the level of methionine was raised to 1.0% the animals gained less weight than the controls and had a lower nitrogen balance. At higher levels of methionine intake, the animals suffered a loss of 40% in body weight, had extremely low nitrogen balances and were found to be devoid of fat. The animals showed increased thyroid size, increased liver size, and increased kidney size on autopsy. The addition of small amounts of arginine to the diet with the methionine greatly improved the situation. The weight gain, nitrogen balance, and organ weights returned toward normal or reached normal limits. The excretion of creatinine increased markedly, indicating that the methyl group of the methionine might be combining with the nitrogen group of the arginine to form creatinine, thus lowering the toxicity of the methionine. This explanation is logical assuming that the methyl group of the methionine is the toxic agent. Addition of glycine to the diet produced the same effect as the addition of arginine, again implicating the methyl group. Possible medical implications were discussed.

JOSEPH W. STRALEY (Physics): *The Infrared and Raman Spectra of a Family of Methane-like Molecules.*

Methane-like molecules include all molecules of the point group T_d which have an atom at its geometric center. All such molecules have four fundamental vibra-

tional modes in common. A determination of the frequencies associated with these modes may yield valuable information regarding the geometry of the molecules and the force constants of the various interatomic bonds. For molecules having such a high degree of symmetry, the problem of determining these frequencies is complicated by the fact that selection rules operate to forbid the observation of certain of these frequencies in either the infrared or the Raman Effect or both. In this case all fundamentals are allowed in the Raman Effect but only the two triply degenerate vibrations are allowed in the infrared.

In the group of molecules CH_4, CD_4, CF_4, $C(CH_3)_4$, $Si(CH_3)_4$ and CCl_4 all of these fundamentals have been observed in either the infrared or the Raman Spectrum. The observation by the author of one of the forbidden infrared bands in methane is explained as being due to a Coriolis interaction with a neighboring allowed frequency. The frequencies observed may be correlated with the mass of the constituent atoms and with their elastic constants.

494TH MEETING, FEBRUARY 12, 1952

JOHN BORDEN GRAHAM (Pathology): *Further Observations on Canine Hemophilia.*

Hemophilic dogs from an inbred strain of Irish Setters have been under observation in our laboratory for five years. The disease in these animals appears identical with that of humans. The genetic defect is transmitted as a sex-linked recessive characteristic, and the animals suffer from abnormal bleeding. By selective breeding, all three of the female genotypes and both male genotypes have been obtained from the original stock.

No evidence of a blood clotting dyscrasia can be found in the heterozygous females, indicating complete recessivity of the characteristic. The homozygous females are bleeders whose defect appears identical with that of the hemophilic males. The mating of bleeder males with bleeder females has produced litters composed exclusively of hemophiliacs.

Blood from the hemophilic animals has been used to devise a semiquantitative assay for the antihemophilic principle of normal dogs (AHF). With this test the plasmas of normal dogs, females heterozygous for hemophilia and normal animals rendered prothrombinopenic by the drug dicumarol or by chloroform intoxication have been shown to have a normal and "steady" level of AHF.

JAN H. R. BEAUJON and WALTER H. HARTUNG (Pharmacy): *A New Tryptophan Analog.*

A procedure was described for synthesis of a new analog of tryptophan, α-aminoskatylmalonic acid. The starting material was diethyl malonate which after nitrosation and catalytic hydrogenation of the resulting diethyl isonitrosomalonate yielded diethyl aminomalonate. The reactive amino function was protected by reaction with benzyl chloroformate. This carbobenzyloxy group was chosen because of the ease of removal by catalytic hydrogenolysis. This prevented any tendency toward mono-decarboxylation of the malonate during the liberation of the amino group.

The diethyl carbobenzyloxyaminomalonate was alkylated with gramine and dimethyl sulfate in the presence of sodium ethoxide. The resulting diethyl carbo-

benzyloxyaminoskatylmalonate was hydrolyzed at room temperature with alcoholic potassium hydroxide solution. Potassium carbobenzyloxyaminoskatylmalonate was formed which was hydrogenated to potassium α-aminoskatylmalonate. This potassium salt was stable, and it was used in the following experiments rather than the unstable free acid.

Rat feeding experiments were carried out. It was found that the analog did not replace L-tryptophan, nor was it antagonistic to it.

Microbiological evaluations seemed to indicate that potassium α-aminoskatylmalonate could not replace L-tryptophan in the culture medium of *L. arabinosus* 17-5. It was also found that the malonate did not inhibit growth of the organism in a complete medium.

495TH MEETING, MARCH 11, 1952

H. G. MCCURDY (Psychology): *Experiments in Group Problem-Solving.*

By an arrangement of interconnected switches (the Group Maze), it is possible to study the behavior and efficiency of small groups in a variety of strictly cooperative problem-solving situations. A program of research utilizing this device was begun at the University of North Carolina in 1949. Experiments conducted thus far have been concerned with the relative problem-solving efficiency of individuals and groups, and with the effect on efficiency of varying the composition and organization of groups. The results may be expressed in a number of different ways, and no brief summary statement can be quite accurate; but in general terms: (1) individuals are more efficient at problem solution than newly-formed groups; (2) slight diminutions in intra-group communication cut down on efficiency; (3) no difference appears between randomly selected groups organized in contrasting fashion as miniature democracies and dictatorships; but (4) when such groups are composed of persons selected by the Frenkel-Brunswik *F*-scale as authoritarian or democratic personalities, the democratic groups composed of authoritarian personalities commit more errors per unit time than the other three combinations. Groups consisting of three individuals only are considered in these particular comparisons, though the apparatus will accommodate groups of six. Experimenters chiefly responsible for the studies indicated under 2, 3, and 4, respectively, were Harold P. VanCott, Wallace Lambert, and Herbert Eber, all graduate students in the Department of Psychology.

J. LOGAN IRVIN and ELINOR MOORE IRVIN (Biological Chemistry and Nutrition): *The Reversible Interaction of Plasma Proteins and Acridine Derivatives.*

The proton-acceptor species of the antimalarials chloroquine (7-chloro-4-(1′-methyl-4′-diethylamino-butylamino)-quinoline and SN-12868 (2-methoxy-6-chloro-9-(1′-methyl-8′-diethylamino-octylamino)-acridine combine reversibly with bovine plasma albumin and with the proteins of fraction III-1 of bovine plasma. This interaction has been evaluated spectrophotometrically with the equation

$$\mathrm{pH} = pK_1' + \log \frac{(D_1 - D)}{(D - D_2) + k_1'[P](D - D_3)}$$

in which pK_1' is the apparent ionization exponent for the group on the aromatic nucleus, k_1' is the association constant for the binding of the first molecule of the quinoline or acridine derivative to a molecule of the protein, $[P]$ is the molar concentration of free protein at equilibrium, and D_1, D_2, D_3, and D are the optical densities at a selected wave length when the quinoline or acridine derivative is completely in the form of the proton-donor species, the free proton-acceptor species, the protein-bound acceptor species, and an intermediate equilibrium state respectively. The association exponents are as follows:

PROTEINS	LOG k'_1 at 30°	
	SN-12868	Chloroquine
Albumin	4.38	3.72
Fraction III-1	4.14	
Fraction II	3.17	

These data have been applied in the description of some of the factors which are concerned in the distribution of these drugs in the animal body. This study also provides a model-system for the evaluation of the interaction of these compounds with enzymatic proteins.

496TH MEETING, APRIL 8, 1952

CARROLL E. WOOD, JR. (BOTANY): *Some Researches on the Biology of Downingia.*

The genus *Downingia* (Campanulaceae) includes about 14 well marked species of small winter annuals occurring primarily in vernal pools in the western United States (especially California) and adapted to a regular seasonal sequence of increasing dryness, rising temperature and lengthening days.

The ranges of several species overlap in part and 2–4 species may sometimes occur together; yet hybrids do not seem to occur in nature. Ecogeographical, seasonal and mechanical differences and differing chromosome numbers (species with 8, 10, 11 and 12 pairs occur) may have operated as reproductive barriers between species, but at present the principal barriers appear to be genetic. More than 1000 attempted crosses between 8 species (representing 3 of 4 well marked species-groups) show that hybrids are difficult or impossible to obtain, even between closely related species, but when obtained are fertile. Seeds either are not set, or abort near maturity or give only an occasional albino lethal, with the exception of *D. insignis* × *D. pulchella* (closely related but distinct species with 22 chromosomes). Here abundant seeds are obtained which germinate as pale green, weak seedlings, many of which die while the remainder flower 1–2 months after the parent species. Pollen fertility is high (92–98%) and back-crosses are vigorous and fertile. An F_2 generation included many inviable combinations (about ½ albinos) but also some vigorous and fertile segregates. The F_1 hybrid, if it occurs, does not appear to survive to flower in nature.

The species investigated are not only distinct morphologically but are natural biological units isolated from each other by strong reproductive barriers.

E. D. PALMATIER (Physics): *Energy Laws of Mesons in Gases.*

497TH MEETING, MAY 13, 1952

LOUIS DUBOSE QUIN (Chemistry): *Some Organic Phosphorous Compounds of Medicinal Interest.*

At the beginning of the meeting Dr. Crockford presented a certificate and a check for $50.00 to Mr. Louis Dubose Quin, as winner of the William Chambers Coker Award in Science for the year 1951–52.

Memorials were read in honor of three members of the Society who died within the past year: Dr. Ralph Walton Bost, Dr. William de Berniere MacNider, and Dr. Ira Winfield Rose. These memorials are printed below.

RALPH WALTON BOST

Dr. Ralph Walton Bost was born in Rockwell, North Carolina, on January 5th, 1901, and was educated at Newberry College and the University of North Carolina, where he received the degree of Ph.D. in 1928. Except for two years when he was an instructor at Tulane University, all of his scientific and professional life was spent at the University of North Carolina, where he started as an instructor, and advanced through the various grades, becoming Smith Professor of Chemistry in 1940 and finally Kenan Professor of Chemistry in the year of his death. He was Acting Dean of the School of Applied Science from 1933 to 1935 and was Head of the Department of Chemistry from 1939.

His chief scientific interests were in the field of organic chemistry. In particular his research activities were concerned with organic sulfur and nitrogen compounds, the relationship between molecular structure and surface activity, reactions in liquid sulfur dioxide, identification of organic compounds, and chemotherapy. In these fields he and his students published over seventy articles; he was also the author, with P. Borgstrom and D. F. Brown, of a comprehensive bibliography of work on organic sulfur compounds. During his years at the University he guided forty M. S. and forty-one Ph.D. candidates. In recognition of his scientific accomplishments he received the Herty Medal in 1950.

He was a long-standing member of the Elisha Mitchell Society, and also belonged to a number of scientific and professional societies, including the Society of Chemical Industry, the Chemical Society (London), the American Physical Society, the North Carolina Academy of Science, Alpha Chi Sigma, Sigma Xi, and the American Chemical Society. In the latter society he held a number of offices. At different times, he was Secretary-Treasurer and Chairman of the North Carolina Section, and he represented the Section in the Council of the national society. He was a member of the Council's Committee on National Meeting and Divisional Activities. He was Secretary of the Division of Organic Chemistry from 1944 to 1949 and was Chairman of the Division in 1950. He was a member of the Board of Directors from 1949. He was on the Organic Advisory Board of Chemical Abstracts and a member of the board of editors of the journal, Organic Chemistry.

RALPH WALTON BOST

He was married to Beulah Cauble on July 12th, 1927. She was a devoted and sympathetic companion, and contributed significantly to the atmosphere which made possible his varied accomplishments.

Dr. Bost was a charter member and officer of the Holy Trinity Lutheran Church of Chapel Hill, and in recognition of his service, the Church has named its library after him.

He was a member of the Chapel Hill Rotary Club, and has been its President.

In the passing of Dr. Bost on September 22nd, 1951, the Elisha Mitchell Scientific Society and the University community have lost one of their most loyal, valued, and distinguished members.

W. H. HARTUNG
R. L. MCKEE
O. K. RICE, *Chairman.*

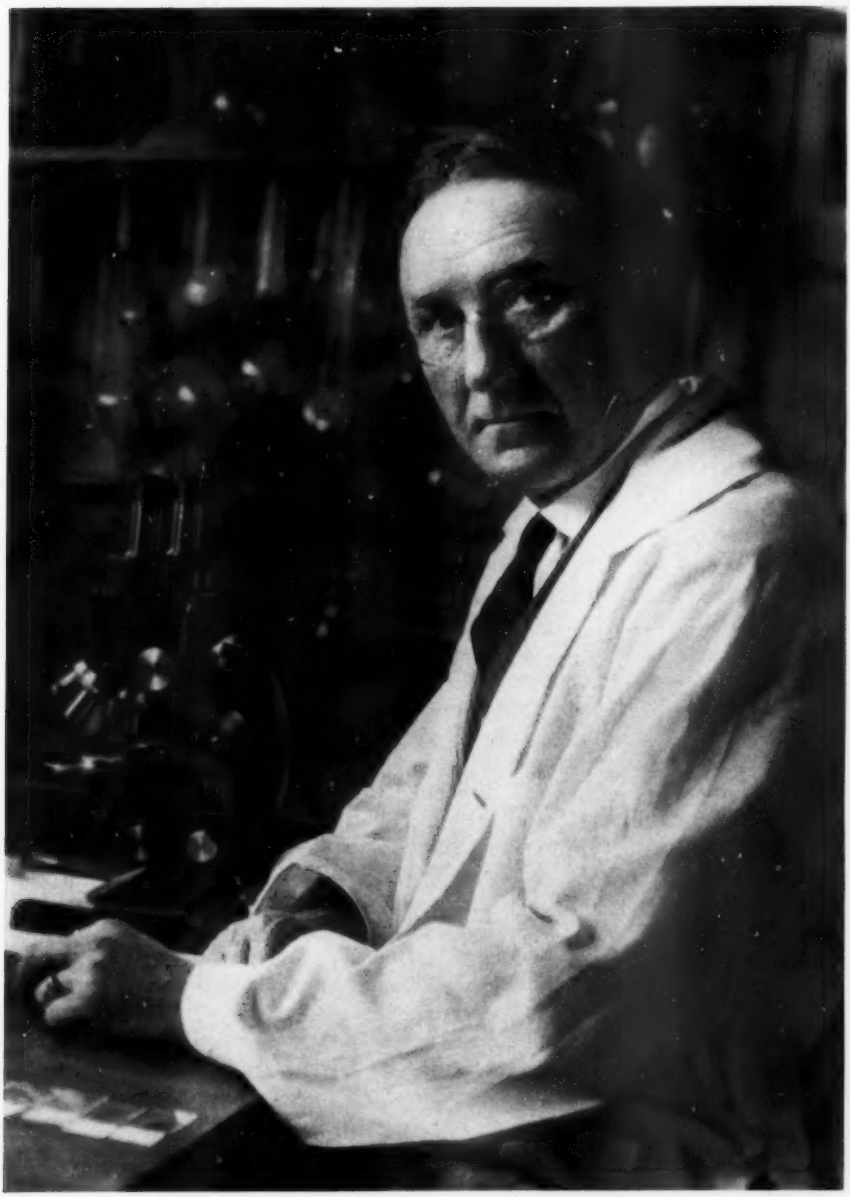
WILLIAM DE BERNIERE MACNIDER

In the passing from this life, May 31, 1951, of William de Berniere MacNider, the Elisha Mitchell Scientific Society recognizes the loss of one of its most faithful and distinguished members.

Dr. MacNider was born in Chapel Hill, North Carolina, on June 25, 1881. He was educated in the local schools and received his college and medical education at the University of North Carolina. His professional education was extended during some subsequent summers through study at the Universities of Chicago, Western Reserve, and Johns Hopkins. Throughout most of his life he was a member of the faculty of this University, being successively Assistant in Biology (1899–1900), Assistant in Anatomy (1900–1902), Assistant in Clinical Diagnosis (1902–1905), and Professor, Kenan professor, and Kenan Research Professor of Pharmacology (1905–1950). He retired as Emeritus Professor in 1950. From 1937 to 1940 he was Dean of the Medical School.

Dr. MacNider was one of the University's most productive scholars. From 1903, at the age of twenty-two when his first paper with H. A. Royster appeared, to his retirement in 1950, over 140 papers were published, most of which concerned the function and pathology of the kidney. His scholarly achievements brought him many honors. He was elected to membership in the National Academy of Sciences, the American Philosophical Society, the American Academy of Arts and Sciences, and many other scientific organizations at home and abroad. He served as president of the American Society for Pharmacology and Experimental Therapeutics, the Society of Experimental Biology and Medicine, the International Anaesthesia Research Society, the Gerontological Society, and the Elisha Mitchell Scientific Society. He was awarded the Gibbs prize of the New York Academy of Medicine, the Research Medal of the Southern Medical Association, aud the Kober Medal of the Association of American Physicians.

Throughout his long career he maintained a keen interest in whatever added to scholarship and the general well-being of the University. His sound judgment and forthright manner of speaking made him a favorite choice of the faculty on elected committees, such as the Advisory Committee, and won for him appoint-



WILLIAM DE BERNIERE MACNIDER

ments of unbroken service on such boards as the Graduate and Library Boards, and such committees as the Grounds and Buildings Committees. Although he gave freely of his time and energy to such committee work, his main interests were in research and teaching. He seldom missed a meeting of the Mitchell Society, and many of his important contributions were first announced before this body. His pertinent comments, in the discussions of papers presented by others before the Society, showed the scope of his knowledge, added to the interest of the meetings, and left with each researcher encouragement and zeal for continued work. One of his last services to the society was to serve on the committee which initiated the establishment of the William Chambers Coker Award.

In his death we have lost an eager scholar, an inspiring teacher, and a devoted friend. To his family, whose loss is greatest of all, we extend our deep sympathy.

W. C. GEORGE
W. R. STRAUGHN
J. N. COUCH, *Chairman.*

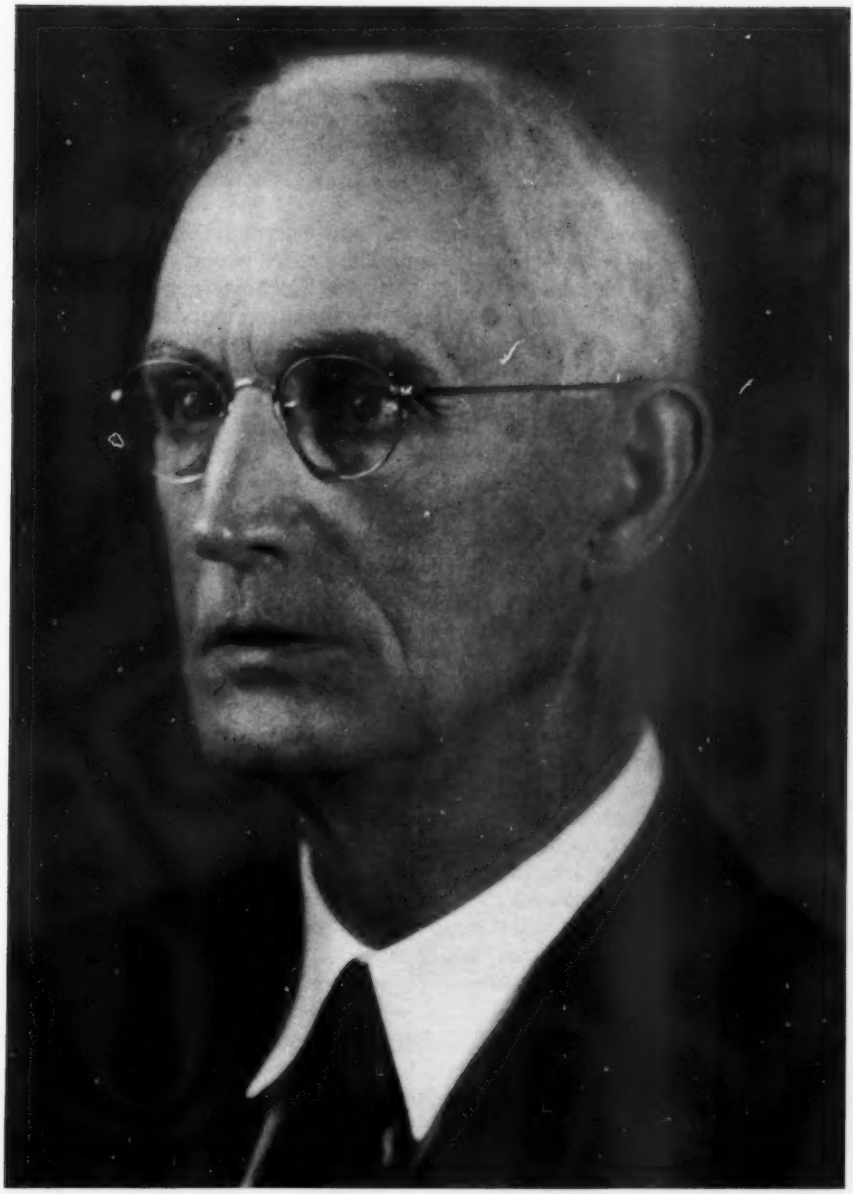
IRA WINFIELD ROSE

Ira Winfield Rose was born at Bentonville, North Carolina, on September 21, 1880 and died on January 14, 1952. He received his early education in the schools of his community, taught in Johnston County from 1898 to 1901, attended Massey Business College in Richmond, Virginia, and in 1902 began his career in pharmacy as a clerk. He received the Ph.D. degree from the University of North Carolina in 1906 and also became a registered pharmacist. He remained ever faithful to his Alma Mater and to his beloved profession.

Mr. Rose established his own professional and successful pharmacy in Rocky Mount. He promptly joined the North Carolina Pharmaceutical Association, an organization that recognized his sterling worth by electing him after four years to the third-vice presidency, a year later to the second-vice presidency, and in 1921 to the presidency. For nearly a quarter of a century he was a member of the North Carolina Board of Pharmacy and he was instrumental in introducing in 1913 the practical laboratory examination as a feature in examining candidates for licensure.

In 1931 his Alma Mater beckoned him, besought him to give up a profitable, professional life to accept the modest fortunes of a professor. Acceptance of this call indicated the measure of the man, increased for him the unqualified respect of his professional brethren, and gave him an unequalled opportunity to become the friend and confidant of twenty graduating classes. His ideals of scholarship, his high principles, unassuming dignity and Christian charity set an excellent example for all. Even after he reached the compulsory age of retirement in 1951 his Alma Mater asked him to continue on an emergency half-time basis. By temperament and disposition Professor Rose was always a teacher, yet on three separate occasions he was asked to serve as Acting Dean of the School of Pharmacy.

Not only was he a teacher, but he also contributed to the advancement of his profession and, while at the University, published in the Carolina Journal of Pharmacy five original papers on dispensing problems.



IRA WINFIELD ROSE

Professor Rose was a member of Kappa Psi fraternity and Rho Chi society and a life member of the American Pharmaceutical Association. He was a regular and faithful member of long standing of the Elisha Mitchell Society. He was a faithful attendant of the Baptist Church.

Professor Rose married Juanita Pearl Penny in 1919, and established a home which was a delight and inspiration for students, colleagues, and a host of friends. Their son, Winfield Penny Rose is following the profession enobled by his father.

In the passing of Ira Winfield Rose the Elisha Mitchell Society has lost a valued and venerable member. Our sympathy is extended to his family.

E. A. BRECHT
FRED SEMENIUK
W. H. HARTUNG, *Chairman.*

Each memorial was approved by a rising vote.

The president presented the Executive Committee's recommendation that Associate Membership dues be raised from $.25 to $.50, and it was unanimously approved.

The treasurer's report was read, and the report of the Auditing Committee (L. L. Garner and J. C. Andrews), saying that the books had been examined and found correct, was read and accepted.

The Nominating Committee (G. O. Doak, A. V. Masket, and D. P. Costello, Chairman) presented the following slate of nominees for the year 1952–53:
President—PAUL E. SHEARIN
Vice-President—C. W. HOOKER

A motion to instruct the secretary to cast a unanimous ballot for this slate was made and unanimously approved.

WAYNE A. BOWERS, *Sec.-Treas.*

GENETICS, TAXONOMY, AND THE RACES OF MAN[1]

By R. Ruggles Gates[2]

Professor Emeritus, King's College
University of London, England

The invitation to speak on this thorny subject left me wondering whether I could honestly discuss this problem, which is both scientific and human, without treading on someone's toes. It is a subject which can be profitably discussed only in an atmosphere of kindly feeling and a complete absence of malice, to which must be added the determination to seek the truth regardless of where it leads. In a subject which has been so deluged with propaganda there will probably be some to dissent, whatever one may say.

I speak as a biologist whose life has been rendered strenuous by the continued effort of a lifetime to keep in touch with the main lines of development during the present century in the diverse fields of botany, cytology, genetics, and experimental zoology, and in the last 28 years in anthropology as well. Occasional ventures have been made into such diverse fields as psychology, with more frequent contributions to taxonomy. Indeed, the problems of species have seldom been far from my thoughts over long periods. One feels that such a sustained effort through the years could only be justified by the presence in the background of some unifying principle, and that principle has of course been evolution. My early work was concerned with the laws and principles of mutation, and in recent years I have been more concerned with the principles of genetics, especially as applied to man. This very brief allusion to what I may call my *curriculum vitae* is merely to indicate that I should not be entirely unfitted to discuss the subject before us.

There have been persistent efforts to decry race, as though there was something unnatural about it. Books have been written to say that race is a "myth", and a dangerous one at that. Others have tried to persuade themselves that race is a "superstition". Someone may yet cap the climax by announcing that race is a disease! In the flight from the idea of race, there have been various attempts to suppress this useful word altogether, as though the word could be altered by fiat. All the Indo-European languages appear to have a word meaning race. Among native tribes, their own name for themselves most frequently turns out to mean "the men". In other words, "we are the men" is generally the somewhat egotistical attitude of all peoples.

Some anthropologists have revived the term ethnic group, first used by Deniker (1913), as an equivalent of race. But many now realize that the term race is too

[1] This paper was given in a symposium on *Genetics and Contemporary Problems* before the North Carolina Academy of Science, Greensboro, N. C., May 2, 1952.

[2] At the time that this paper was given, the writer was an Honorary Research Fellow in the Department of Anthropology at Harvard University, Cambridge, Massachusetts, U. S. A.

deeply embedded in language to be done away with, even if this were desirable. Surely it is better to recognize that races exist, and will continue to exist, whether we like it or not. The problems connected with race are not solved by denying their existence. The term is of course not confined to man, but it is continually being used by botanists and zoologists in reference to subspecific groupings.

It has often been objected that the term race can be used in a more inclusive or a more restricted sense. This is indeed one of its advantages. Thus we may speak of the human race, the Mongol race, or the Eskimo race. But no one is in doubt as to the meaning, although the Eskimos can be included in the Mongolian race, and the Mongols in the human race. This flexibility of the term is a distinct advantage, which is not shared by the term species, unless we speak of subspecies or microspecies as units within a species, which is not always feasible.

The word race thus has much in common with the term species. As pointed out elsewhere (Gates, 1951), it is impossible to frame a universal definition of species which will apply in all groups of organisms. Similarly, race is used in various restricted or inclusive senses, and the meaning is generally clear from the context. Montagu (1951) has recently made a formulation which appears to be acceptable. "A species is a group of populations. The group is the species, the populations are the races." But this of course, and of necessity, leaves undetermined what are the limits of the species or the race in any particular case. In the last analysis, the determination of the limits of a species, subspecies, race or other taxonomic category is a matter of knowledge, judgment, convenience and consistency in classifying the organisms found in any particular phylum or group.

Stern (1949, p. 558) defines race as: "A genetically more or less isolated division of mankind possessing a corporate genic content which differs from that of all other similar isolates". Montagu complains that this fails to distinguish race from species. That, in my view, is its merit, the distinction being one of degree and not of kind. On the other hand, the "degree of actual or potential isolation", cited by Montagu, should not be regarded as a necessary quality of species.

Of course, it would simplify matters greatly if some rule-of-thumb method could be applied as the universal criterion of species. Mayr (1942) has strongly advocated intersterility or reproductive isolation as such a criterion, but this leads to bizarre and inconsistent results as between different groups of animals. It produces the extreme lumping of species in some groups (including man, as we shall see) and in others the recognition of "species" with practically no phenotypic or morphological difference at all.

Those who have described species of living plants or animals consider that their task is done when they have not only delimited the characters of a species in relation to those of other species, but also have indicated how it may vary from one population to another. These neontologists may know nothing of the history of their species except in so far as this may be indicated by its geographical distribution in comparison with related species.

By contrast, the paleontological taxonomer must perforce take into account time as a third dimension in the delimitation of his species. Generally a break in the strata, or an unconformity, or a failure of fossilization at some point in

the particular lineage he is studying, will relieve him of the necessity to decide where the variations in his line of descent are large enough to justify delimiting any species from an ancestral or a descendant species. Only in relatively rare cases must he exercise his judgment as to how much of a line of descent should be broken off, as it were, like a piece from a bar of candy, and named as a distinct species. In the fossil record of evolution there are few natural joints, apart from the artificial geological gaps already mentioned, to indicate to the naturalist where one species has ended and another begun. Nature continues her way regardless of any difficulties the biologist may encounter. Although for the paleontologist the necessity of drawing an artificial line may be rare owing to the incompleteness of the geological record, yet it should never be overlooked by any biologist that there has always been complete breeding continuity between the species he is describing and an ancestral species. Although this continuity may be partly broken by mutations, yet these breaks may never be great enough to prevent a continuous line of breeding descent. The only exception to this is in cases of amphidiploidy, which occur chiefly in plants. The modern work in genetics, as well as in paleontology, confirms the earlier views of Darwin that the transition from a particular species to a descendant species has generally been a gradual one, always marked by breeding continuity. Whether the origin of certain phyla and orders of animals may have been more abrupt need not concern us here. But even in the most abrupt transition the new species must have had immediate ancestors, although placed in another species.

Elsewhere (Gates, 1951) I have developed the theme that intersterility between contemporary species of plants or animals is an incidental condition which may or may not arise in the evolution of species. In some groups intersterility may arise between forms which show little or no phenotypic difference. This does not confer upon them the rank of species. At the other extreme are cases both in plants and animals in which a large amount of evolution has occurred without the development of intersterility. Examples in plants are *Oenothera*, *Catalpa* and *Platanus*. The Eastern and Western plane tree, for example, must have evolved separately for millions of years, yet they are perfectly interfertile.

Among animals, perhaps man himself is as good an example as any, of wide interfertility. This applies not only to the primary races or species of modern man (which Darwin regarded as subspecies), but the fossil evidence indicates that species as distinct as Neanderthal and CroMagnon man intercrossed and produced fertile offspring. There is much other evidence suggesting wide interfertility between contemporary species of fossil man. This is in strong contrast with such insect genera as *Drosophila*, in which forms showing practically no phenotypic difference may nevertheless be intersterile. A discussion of the reasons for these contrasts in interfertility is reserved for another occasion. Here it need only be remarked that a sound taxonomy must be based on morphological difference rather than on the sporadic occurrence of intersterility. Hooton (1946, p. 447) says, "All existing forms of man are usually included in one species, *Homo sapiens*, although the differences between the several races are quite as marked as usually serve to distinguish species in other animals."

It is particularly gratifying to find a paleontologist of the eminence of Dr. Simpson (1951) forcibly expressing the same views as mine regarding the two horns of the intersterility dilemma. He says (p. 59), "Absence of actual interbreeding between two populations is not, even in theory, a sufficient criterion that they are distinct species unless there is evidence that this does in fact make them separate phyla,[3] with different evolutionary destinies or potentialities." His qualification, "unless . . .", seems unnecessary, since it could only be determined by coming back here some ten thousand years hence to see what has happened. Species are described as they are now and not as they may be conjectured to become at some future time.

On the other horn of the intersterility dilemma we may again quote Simpson (p. 59): "All biologists know that species, distinct and valid by genetic as well as by all other usual modern definitions, may still be richly interfertile." This would seem to imply that he has converted some of his colleagues, and it is very satisfactory as supporting completely my position regarding the relations between species and sterility.

Having been thus explicit in denying that intersterility can be used as the criterion of species, it is a little disconcerting to find a biologist with Simpson's acuteness of mind partly reversing himself with the statement: "To virtually all modern biologists, the word 'species' has the (latter) meaning" of "a phylum evolving independently from all others". On the contrary, to virtually all botanists and to many zoologists the term species implies a certain amount of morphological difference. In the great bulk of described and recognized species it is not known, and is not essential to know, whether they do or can cross in nature and produce fertile hybrids.

As a paleontologist, Simpson naturally emphasizes the time element in relation to species. In the cases such as I have cited, in which species which have been separated for a million years may be fully fertile when brought together, no botanist, so far as I am aware, would think for a moment of calling them one species on that account. Such cases show the absurdity of regarding the interbreeding behavior as the fundamental criterion of species. It is surprising that so acute a thinker as Simpson should have overlooked the significance of the inescapable fact, which is particularly obvious in paleontology, that continuous reproduction precludes the possibility of intersterility between a species and its descendant species. If all animals are derived from one original ancestor then, on a sterility basis, they are all one species, for each has been produced by breeding from an ancestral species. This shows how absurd is the intersterility criterion as applied in theory to species, especially paleontological ones.

The results of applying this criterion to the evolution of man, together with a desire to minimize the differences between human races, leads Mayr (1951) to a classification of fossil hominids which can only be characterized as fantastic, inconsistent, and absurd. Having defined a species as "a group of actually or potentially interbreeding natural populations that is reproductively isolated from

[3] This use of the term phylum seems rather unfortunate, as it is quite different from the general usage in zoology.

other such groups", he should have drawn the logical conclusion from his premises, that all hominids, living and fossil, are one species. Finding this conclusion beyond the power of his auditors to swallow, he announces instead that "never more than one species of man existed on the earth at any one time". Yet he admits, as Sir Arthur Keith (1948) has emphasized, that "primitive man was much more broken up into small scattered tribes with little contact with each other, intensely subject to local selective factors".

To continue Mayr's excursus into anthropoid and human taxonomy, the chimpanzee and gorilla are combined into one genus, despite their great morphological differences. He finds no justification for placing anthropoids and hominids (which he reduces to the single genera *Pongo* and *Homo*) in separate families, and "there is even less justification for placing South African man in a separate subfamily, the Australopithecinae". Five species in three genera may be a generous taxonomic allowance for this group of South African ape-men, although it is defended by Broom (1950), who is a leading authority in paleontology. But Mayr reduces them all to one species, not of Australopithecus but of *Homo*——*H. transvaalensis*! Java and Peking man are another species, *Homo erectus*. *Meganthropus* and *Gigantopithecus* are unplaced (so perhaps there may have been two or three contemporaneous species of man after all). All others, including modern man, CroMagnon and all the Neanderthaloids, are relegated to *Homo sapiens*. Thus the whole of the evolution of human and sub-human types scattered over the earth's surface for a million years is reduced to one genus with three species!

The confusion of mind involved is patent when Mayr speaks of the Australopithecines as "man" and *Pithecanthropus* as "ape-man"; but he has done a real service in showing the logical result of his taxonomic ideas as applied to man.

In spite of all this, Mayr admits that if the bones of the Congo pygmies and the Watusi, a Negro people with whom they are in contact, were found by a paleontologist a million years hence, "he might well think that they belonged to two different species". He expresses the opinion that man "cannot speciate" because he has occupied all the ecological niches on earth. This may perhaps be true of modern man with his huge populations. But his sparse occupation of temperate and northern zones, which took place at a relatively late date, was only made possible by his invention of clothing and habitations, or the use of caves as a protection against the climate. Clothing was only adopted perforce by northern man as a result of the refrigeration during the Pleistocene. By that time most of man's differential evolution had been completed, and the convergence of the types already created under comparative isolation had begun. This convergence has gone on with increasing pace ever since, by increases in population bringing the various types into closer contact. With the invention of agriculture *ca.* 8000 B.C. (Keith 1948) this process was greatly accelerated. (The latest radiocarbon date for the Jarmo preceramic village site in Kurdistan is 6707 ± 320 years.)

It is obvious that Neanderthal man could not have survived the onset of the Ice Age in Europe without the invention of clothing. That Pleistocene man was

more variable than his modern descendants is indicated by the skulls from the upper cave at Choukoutien and the skeletons from Mount Carmel.

Among the questions asked at the end of this exhibition of superlumping was one by Dr. Lasker, who asked how Mayr could justify placing man, gorilla and orang in three separate genera, while man, the Australopithecinae, and Piltdown were all crowded into one genus. Such questions serve to expose the inconsistencies of the whole scheme. Such confused thinking about race often begins with denial of one's own race.

The conclusion appears to be now widely accepted that the Australopithecinae of South Africa, known through the work of Dart, Broom and others, represent a starting point for humankind. Their relationship to the more anthropoid Dryopithecinae in the Siwalik Hills of Northern India remains obscure, but it is quite possible that such genera as *Ramapithecus*, with humanoid dentition, may prove to be closely related to or even within the group of Australopithecinae. These and many other ancestral forms have been discussed elsewhere (Gates, 1948). The South African group alone, with their ancestors, should be quite capable of producing all the later genera and species of mankind. In this sense man's ancestry is probably monophyletic, but we must assume that the group spread widely from South Africa and perhaps from India to evolve later the two species of *Pithecanthropus* in Java and Northern China, at a higher level of human development. Broom (1950) has stated that the *Meganthropus* jaw from Java, the largest human jaw ever found, is equalled in size by an undescribed Australopithecine jaw found in South Africa. He suggests that the three huge molar teeth (*Gigantopithecus*) of unknown age from caves in Southern China may belong to a gigantic Asiatic member of this South African group.

We may turn now for a moment to the blood groups. Their inheritance as single units in all races of mankind, whereas nearly all other racial differences in modern man depend on multiple genes or polygenes, gives them great value as differentiating characters additional to those of morphology. Anthropologists have been slow in adopting the blood groups into their thinking on matters of racial relationships and human phylogeny. While they sometimes serve most usefully in limiting the time at which certain migrations could have taken place, as pointed out many years ago in the case of American Indians and Eskimos (Gates and Darby, 1934), in other cases, as in parts of Africa, they may appear to conflict with conclusions based on other lines of evidence. Further knowledge will no doubt ultimately eliminate any such conflict.

In a recent discussion of some of the later results of blood grouping in relation to race, Mourant (1951) shows how illuminating these results can be. For instance, the Basques differ from other European peoples both in the ABO and the Rh allele frequencies. This strongly supports the evidence from language (Basque being the only non-Indo-European language in Western Europe), from tradition and from physique, that the Basques, who still survive in parts of France and Spain around the Bay of Biscay, have mixed very little with other peoples. They perhaps date from the Paleolithic, and Vallois (1945) shows that their characteristic blood grouping is still found widely in the southeast population of modern

France. The Lapps of Northern Europe show various resemblances to the Basques, including a low percentage of the B blood group. If they should prove, like the Basques, to have a high Rh frequency, then they might belong to the same early stratum of population.

In Catalonia, Sardinia and parts of Italy the Rh analysis likewise gives clear evidence regarding the origins and history of the population, conclusions which can frequently be verified or supported by history. The high frequency of the cDe (Ro) allele in Negroes is a means of tracing Negro ancestry in populations of Southern Europe.

In the same way, Mourant shows that the blood group picture of recent Jewish immigrants from Eastern Europe to Western Canada is still typical of the Eastern Mediterranean where they originated. He also finds that they have a higher frequency of cDe than anywhere in Europe except Galicia (northwestern Spain), and he attributes this to African blood picked up during the sojourn of the Israelites in Egypt. The Jewish race is thus basically typical of the Eastern Mediterranean in blood groups as well as in physique.

In a further analysis of Mediterranean peoples, Mourant is able to draw certain conclusions regarding thalassemia, a hereditary (dominant) condition of the blood, which produces Cooley's anaemia and is known to be found mainly in Mediterranean populations. He finds a high frequency of thalassemia in regions such as Sardinia and Ferrara (in N. Italy) where the cDe (R_1) blood group also has a high frequency. Mourant disclaims the use of the term "Mediterranean race" for the peoples around the Mediterranean Sea, as belonging to the "older anthropology"; but the evidence of history and protohistory shows that many parts of the Central and Western Mediterranean were colonized and peopled from the Eastern Mediterranean, i.e., by men typical of the Mediterranean race, which is therefore a reality though not a unity (see Sergi, 1914).

I have been criticized for suggesting (in *Human Ancestry*) that the primary races of living man are the equivalent of species, and would be considered as such if we were dealing with monkeys, anthropoids or other animals. However, I am by no means the first to make this suggestion. Later developments, which have been considered elsewhere (Gates, 1952), serve to emphasize the reasonableness of this view. But man has always been reluctant to view himself objectively and dispassionately. In a stimulating discussion of racial characters, their origin and adaptiveness, Coon, Garn and Birdsell (1950) suggest that modern mankind has developed from six "stocks". These I should call species, but as they point out, the term species is relative. As we have seen, there is no hard and fast rule, such as intersterility, which can be invoked to avoid morphological considerations.

There is another aspect of racial relationships which I only have time to touch upon briefly. The taxonomy of the races of mankind is a matter on which biologists may differ. It is an academic matter which does not affect our personal attitude to individuals of other races. I speak as one who has been in contact with native peoples in many parts of the world, as one who is interested in the study of inheritance in all mankind. One's own self-respect necessitates the treat-

ment of all men of every race as worthy according to their aims and their achievements. Keith, in his admirable book (1948) shows how, in human evolution, "co-operation was combined with competition". Herbert Spencer showed how every primitive human group has its code of amity (to its own members) and its code of enmity (to all others). When different races live together in the modern world they must learn to apply the code of amity to each other and to suppress the code of enmity. The modern world has many areas where these rules of ethics need to be applied. Three of the world's primary races or species—Whites, Negroes and Indians—occupy this country in large numbers. The experience of other countries does not lead one to favor inter-racial crossing as a solution of the problems involved. Intermixture of these races has already gone far enough to produce what is in effect a third or intermediate race; and as a result of Mendelian segregation thousands pass for white every year. The colored people naturally show every possible combination of the many physical and mental genetic differences between the parent races. Many of these are bound to be misfits as regards their particular combination of mental and physical differences. The mental conflict in some such cases is pitiful to behold. Surely it is better not to multiply such cases by further crosses. Among some Indian tribes in particular the feeling against miscegenation appears to be strong. The fostering of this feeling by both Blacks and Whites, together with application of the ethics of amity, appears to give the best solution of this difficult question.

There is another aspect of human evolution which I would like to touch upon here, namely that evolution has not always taken an upward course; and that some human evolution, like many lines of animal and plant evolution, has been in a downward or degenerate direction. In certain cases it has led to gerontomorphism, as in some of the Neanderthal men and, according to some, in the Australian aborigines. Broom (Broom and Schepers, 1946) and others have directed attention to the pedomorphism in the Bushmen of South Africa. The Boskop race, based on a skull and several skeletons found in the Transvaal and named by Broom *Homo capensis*, had a huge head and might have been ancestral to both CroMagnon and Neanderthal man. The modern descendants of Boskop man include the Hottentots and the Bushmen. As Broom says (p. 264), "The pre-Bushman races possessed brains much larger than ever found among the world's known geniuses. Today the descendants of these phenomenal ancestors have brains on an average smaller than those found amongst other living members of the species *Homo sapiens*". Boskop man "had larger brains than they could usefully employ according to our conception of the nature of the functions of the brain". This huge type of head still appears occasionally among the modern Hottentots, but the method of its inheritance is unknown. At any rate the modern Bushmen of the Kalahari desert are but degenerate representatives of this former majestic physical type.

The Boskop race and their descendants in South Africa may serve as an example of what can happen to a race, even of the finest physique. History is strewn with races which have had their flowering time and then degenerated. Many causes have been suggested, such as racial crossing, depletion of the leading

stocks through war, or endemic diseases such as malaria. I shall not attempt to discuss them here, but will merely point out that, whatever the factors and conditions involved, the ultimate cause was reversed selection, in which the less endowed elements of the population outbred the better endowed.

In the discussion of genetics and disease we have heard something about the effects of deleterious mutations on the race. The subject has recently been discussed at length by Muller (1950), who points out how the relaxation of natural selection under conditions of civilization necessarily increases the load of mutations carried in the germ plasm of the population. This is not an argument that the medical profession should slacken its efforts to cure disease, whether they be hereditary or otherwise; but it does lay a responsibility upon all medical men to know the somewhat complicated laws of inheritance of hereditary diseases and abnormalities, and to advise their patients against the uncurbed reproduction of serious physical and mental defects.

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BEAM DEFLECTION WHEN THE MOMENT OF INERTIA IS VARIABLE*

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If bending forces are applied to a beam, say AB in Fig. 1, each elementary segment will rotate through an angle $\Delta\theta$ as expressed by

$$\Delta\theta = \frac{M\Delta s}{EI}, \tag{1}$$

in which

M = bending moment (usually in inch-pounds);
Δs = length of elementary segment;
E = modulus of elasticity of the material;
I = moment of inertia of the cross-sectional area about the centroidal axis.

As a check on the validity of Eq. (1), it follows that the product $M\Delta s$ properly belongs in the numerator while EI is in the denominator. This is true since angular rotation increases with the amount of bending and the length, and decreases with stiffness of the material and size of the cross section.

The deflection of the beam when loaded vertically must be small to keep the stresses within allowable limits, hence Δx may be substituted for Δs without appreciable error.

Since the angle θ_{ba} (greatly exaggerated) between tangents at A and B, Fig. 1, is the sum of the angles between tangents at the ends of all segments Δx from A to B, we may write

$$\theta_{ba} = \int_A^B \frac{M\,dx}{EI} = \int_0^L \frac{M\,dx}{EI}. \tag{2}$$

Also, since y_{ba} is the sum of the portions Δy corresponding to all segments Δx from A to B, we may write

$$y_{ba} = \int_0^L (L - x)\theta_{ba} = \int_0^L \frac{M(L - x)\,dx}{EI} \tag{3}$$

It follows that the expressions in Eqs. (2) and (3) have geometric meanings so that in the ordinary case of loaded beams it is easier to use these geometric forms than to carry out the integrations.

Thus if M be plotted as ordinate for all values of x between A and B as if I were constant, then Eq. (2) represents the value of the area under the moment curve between A and B, and Eq. (3) represents the moment about B of

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the area of the moment curve between A and B. In case the moment of inertia is not constant (the case primarily under consideration), the curve M/EI will be used instead of the curve M, but the reasoning would be similar throughout. Hence

$$\int_0^L \frac{M\,dx}{EI} = \sum_0^L \frac{F}{EI}$$

and

$$\int_0^L M(L-x)\,dx = \sum_0^L \frac{F\bar{x}}{EI},$$

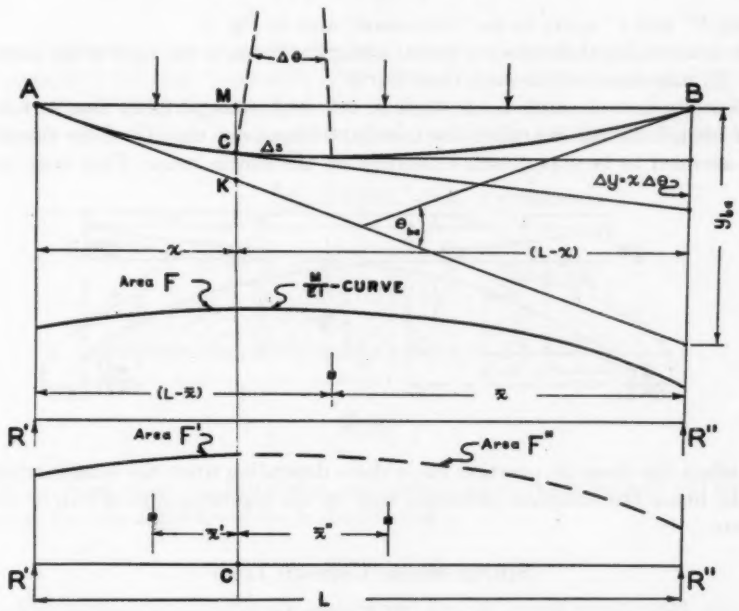

FIG. 1.

in which F and $\bar{x}$ denote respectively areas under the bending moment curves and distances from their centroids to point B.

In Fig. 1, the deflection y, at any point C, from the straight line AB is given by

$$y = KM - KC = \frac{x}{L}\,y_{ba} - y_{ca} \tag{4}$$

Hence

$$EIy = \frac{x}{L}\,F\bar{x} - F'\bar{x}', \tag{5}$$

where F, F', $\bar{x}$, and $\bar{x}'$ are indicated in Fig. (1).

Now if the moment-area ordinates between A and B be regarded as a continuous loading on the beam supported at A and B, counting one unit of weight for each unit of area, the left-hand reaction at A, called R', due to this imaginary loading is $(F\bar{x}/L)$, and therefore

$$EIy = R'\bar{x} - F'\bar{x}'. \tag{6}$$

In similar fashion, if R'' is the right-hand reaction, at B, due to the fictitious M/EI-loading on this so-called "Conjugate Beam", we have

$$EIy = R''(L - \bar{x}) - F''\bar{x}'', \tag{7}$$

where F'' and $\bar{x}''$ apply to the "uncrossed" area in Fig. 1.

In determining deflections of points along the beam to the *right* of the center, Eq. (7) may involve less work than Eq. (6).

Consider now a simple beam, that is, one with ends perfectly free to rotate as if hinged. Actual restraints due to adjacent spans or rigidity of the supports are assumed to be superposed separately on the simple beam. They may tend

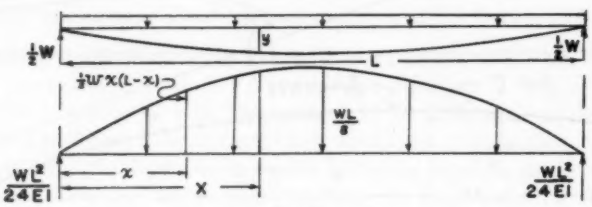

FIG. 2.

to deflect the beam in question *up* or *down* depending upon the combination of loads, hence the resultant deflection may be the algebraic sum of two or three effects.

SIMPLE BEAM: UNIFORM LOAD

(a) *EI Constant*

Let the simple beam of span L ft carry a uniform load of w lbs per ft, or a total load of $W(=wL)$, as indicated in Fig. 2.

The moment curve (I constant) is a parabola with maximum ordinate (at the center) $= \frac{1}{8}wL^2 = \frac{1}{8}WL$.

At any section x ft from the left end, in Fig. 2a, the ordinate to the moment curve is $\frac{1}{2}wx(L - x)$, which becomes $\frac{1}{8}wL^2$ when $x = \frac{1}{2}L$.

Making use of the Conjugate Beam principle, the end reactions to the beam under the M/EI-loading become

$$R' = R'' = \tfrac{1}{2}F = \tfrac{1}{24}\frac{wL^3}{EI} = \tfrac{1}{24}\frac{WL^2}{EI}.$$

Then applying Eq. (6) for the deflection y at any section X ft from the left end, we get

$$EIy = \tfrac{1}{24}wL^3X - \tfrac{1}{2}w\int_0^X x(L-x)(X-x)\,dx$$

$$= \tfrac{1}{24}wL^4\left[\frac{X}{L} - 2\left(\frac{X}{L}\right)^3 + \left(\frac{X}{L}\right)^4\right].$$

Hence

$$y = \frac{CWL^3}{EI}, \tag{8}$$

where C represents the expression in the bracket multiplied by $\tfrac{1}{24}$.

Values of C for each hundredth interval of X/L have been compiled by the author. Hence using Eq. (8), with W, L, E, and I known, the deflection of a uniformly loaded beam can be determined readily for 100 different positions.

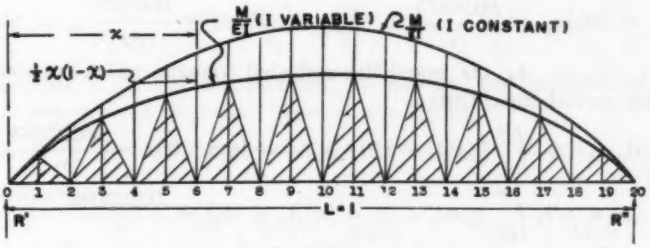

FIG. 3.

Since the beam itself is symmetrical, the deflections at the right half will equal those at corresponding positions in the left half.

(b) *EI Variable*

In Fig. 3, the load is uniformly distributed but the beam may or may not be symmetrical, since I is variable. Deflection coefficients will now be determined on the basis of the unit beam, where $L = 1$, $W = 1$, $E = 1$, and $I = 1$. The actual deflection is found by multiplying the appropriate coefficient by WL^3/EI, in which I will usually be taken as I(minimum).

For the unit beam, the ordinate to the M/EI-curve, distant x from the left end, becomes $\tfrac{1}{2}x(1-x)$; otherwise it is divided by r, the ratio of the actual I at that section to I(minimum).

Let the unit beam be divided into 20 equal parts each of length 0.05; and at sections through points 1, 2, 3, etc, if $I_0 = I$(minimum), let $r_1 = I_1/I_0$, $r_2 = I_2/I_0$, $r_3 = I_3/I_0$, etc, where I_1, I_2, I_3, etc, equal the actual moment of inertia of the corresponding sections.

Next, let m_1, m_2, m_3, etc, equal the actual ordinates at points 1, 2, 3, etc; and noting that the ordinates to the unit M/EI-curve $= \frac{1}{2}x(1-x)$, we have

$$m_1 = \tfrac{1}{2}\frac{(.05)(.95)}{r_1} = \frac{.02375}{r_1}, \qquad m_2 = \tfrac{1}{2}\frac{(.10)(.90)}{r_2} = \frac{.045}{r_2},$$

$$m_3 = \tfrac{1}{2}\frac{(.15)(.85)}{r_3} = \frac{.06375}{r_3}, \qquad \text{etc.}$$

The area under the M/EI-curve is divided into triangles, as indicated in Fig. 3, where the shaded areas, left to right, are denoted by A_1, A_3, A_5, etc. These triangles are bounded by straight lines except for the left part of A_1 and the right part of A_{19}. It follows that

$$A_1 = 1/r_1 \int_0^{.05} \tfrac{1}{2}x(1-x) + \tfrac{1}{2}m_1(.05) = \frac{.0011979}{r_1};$$

$$A_3 = (.05)m_3 = \frac{.0031875}{r_3}; \qquad A_5 = (.05)m_5 = \frac{.0046875}{r_5};$$

$$A_7 = (.05)m_7 = \frac{.0056875}{r_7}; \qquad A_9 = (.05)m_9 = \frac{.0061875}{r_9}; \qquad \text{etc.}$$

Let A_2, A_4, A_7, A_8, etc, equal the unshaded triangles which are bounded at the top by curved lines. Then

$$A_2 = 1/r_2 \int_{.05}^{.15} \tfrac{1}{2}x(1-x) - \tfrac{1}{2}A_3 - \tfrac{1}{2}m_1(.05) = \frac{.00227083}{r_2};$$

$$A_4 = 1/r_4 \int_{.15}^{.25} \tfrac{1}{2}x(1-x) - \tfrac{1}{2}(A_3 + A_5) = \frac{.00402083}{r_4};$$

$$A_6 = 1/r_6 \int_{.25}^{.35} \tfrac{1}{2}x(1-x) - \tfrac{1}{2}(A_5 + A_7) = \frac{.00527083}{r_6};$$

$$A_8 = 1/r_8 \int_{.35}^{.45} \tfrac{1}{2}x(1-x) - \tfrac{1}{2}(A_7 + A_9) = \frac{.00602083}{r_8};$$

$$A_{10} = 2/r_{10}\left[\int_{.45}^{.50} \tfrac{1}{2}x(1-x) - \tfrac{1}{2}A_9\right] = \frac{.00627082}{r_{10}}; \qquad \text{etc.}$$

Summing up the areas A_1, A_2, A_3, etc, we have

$$F = \frac{1}{10{,}000}\left[\frac{11.98}{r_1} + \frac{22.71}{r_2} + \frac{31.87}{r_3} + \frac{40.21}{r_4} + \frac{46.87}{r_5} + \frac{52.71}{r_6} + \frac{56.87}{r_7} + \frac{60.21}{r_8} + \frac{61.88}{r_9} + \frac{62.71}{r_{10}} + \frac{61.88}{r_{11}} + \cdots \frac{11.98}{r_{19}}\right]. \tag{9}$$

If the member is symmetrical, $r_{19} = r_1$, $r_{18} = r_2$, $r_{17} = r_3$, etc., then Eq. (9) becomes

$$F = \frac{1}{10{,}000}\left[\frac{23.96}{r_1} + \frac{45.42}{r_2} + \frac{63.74}{r_3} + \frac{80.42}{r_4} + \frac{93.74}{r_5} + \frac{105.42}{r_6} + \frac{113.74}{r_7} + \frac{120.42}{r_8} + \frac{123.76}{r_9} + \frac{62.71}{r_{10}}\right]. \tag{10}$$

If the $r's = 1$ (I constant), Eq. (10) reduces to $\frac{1}{10{,}000}[833.33] = 1/12$, which is a check, since the area under a parabolic curve equals $2/3 \times$ (the circumscribed rectangle) $= 2/3 \times 1 \times 1/8 = 1/12$.

To find the left-hand reaction R' to the unit conjugate beam (Fig. 3), we apply $F\bar{x} = A_1 \times .95 + A_2 \times .90 + A_3 \times .85 + \cdots + A_{19} \times .05$, which reduces to

$$R' = \frac{1}{10{,}000}\left[\frac{11.38}{r_1} + \frac{20.44}{r_2} + \frac{27.09}{r_3} + \frac{32.17}{r_4} + \cdots \frac{0.60}{r_{19}}\right] \tag{11}$$

If the $r's = 1$ (I constant),

$$R' = \frac{1}{10{,}000}[416.67] = 1/24, \quad \text{which is a check, since} \quad 1/12 \times 1/2 = 1/24$$

Likewise the right-hand reaction $R'' = F - F\bar{x} = A_1 \times .05 + A_2 \times .10 + A_3 \times .15 + A_4 \times .20 + \cdots A_{19} \times .95$, which leads to

$$R'' = \frac{1}{10{,}000}\left[\frac{0.60}{r_1} + \frac{2.27}{r_2} + \frac{4.78}{r_3} + \cdots + \frac{20.44}{r_{18}} + \frac{11.38}{r_{19}}\right]. \tag{12}$$

Substituting in Eq. (6), and reducing, we may find the deflection at any one of the 20 points along the beam. Thus, at the 4th pt,

$$y_4 = R' \times .20 - A_1 \times .15 - A_2 \times .10 - A_3 \times .05 - 1/2 A_4 \times .05/3;$$

which reduces to

$$y_4 = \frac{1}{10{,}000}\left[\frac{0.479}{r_1} + \frac{1.817}{r_2} + \frac{3.824}{r_3} + \cdots \frac{0.454}{r_{18}} + \frac{0.120}{r_{19}}\right] \tag{13}$$

If the $r's = 1$, $y_4 = \frac{1}{10{,}000}[77.33]$, and the actual deflection is

$$y_4 = (.007733)\frac{WL^3}{EI}. \tag{14}$$

For the special case (not unusual) when the member is symmetrical, $y_{19} = y_1$, $y_{18} = y_2$, etc, then we have

$$y_4 = \frac{1}{10{,}000}\left[\frac{0.60}{r_1} + \frac{2.27}{r_2} + \frac{4.78}{r_3} + \frac{7.71}{r_4} + \frac{9.37}{r_5} + \frac{10.54}{r_6} + \frac{11.37}{r_7} + \frac{12.04}{r_8} + \frac{12.38}{r_9} + \frac{6.27}{r_{10}}\right] \tag{15}$$

Table A gives deflection coefficients applying to each of the 20 points of equal division along symmetrical beams. This table represents in effect 10 different formulas, one of which is Eq. (15). The first column indicates the subscript of y (the section); the first row gives the subscript of r; and the tabular quantities are the numerators of the several fractions. The quantities in the last column apply to the special case of beams with constant section throughout, where $r_1 = r_2 = r_3$, etc $= 1$.

Deflections under other loadings such as concentrated loads and various load combinations on the span, have been treated in similar fashion.

TABLE A

Deflection Coefficients for Symmetrical Beam Uniformly Loaded

	1	2	3	4	5	6	7	8	9	10	SUM IF $r'_8 = 1$
1	0.57	1.14	1.59	2.01	2.34	2.63	2.84	3.01	3.09	1.57	20.7
2	0.60	2.08	3.19	4.02	4.69	5.27	3.69	6.02	6.19	3.14	40.9
3	0.60	2.27	4.51	6.03	7.03	7.91	8.53	9.03	9.28	4.70	59.9
4	0.60	2.27	4.78	7.71	9.37	10.54	11.37	12.04	12.38	6.27	77.3
5	0.60	2.27	4.78	8.04	11.33	13.18	14.22	15.05	15.47	7.84	92.8
6	0.60	2.27	4.78	8.04	11.72	15.37	17.06	18.06	18.58	9.41	105.9
7	0.60	2.27	4.78	8.04	11.72	15.81	19.43	21.07	21.66	10.98	116.4
8	0.60	2.27	4.78	8.04	11.72	15.81	19.90	23.58	24.75	12.54	124.0
9	0.60	2.27	4.78	8.04	11.72	15.81	19.90	24.08	27.33	14.11	128.6
10	0.60	2.27	4.78	8.04	11.72	15.81	19.90	24.08	27.85	15.16	130.2

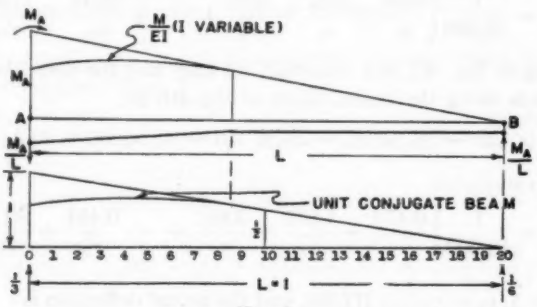

FIG. 4.

END RESTRAINING MOMENTS

In a continuous beam arrangement constructed monolithically the deflection caused by loads on other spans to the right and left of the member has been evaluated for each twentieth point. Thus the upward (or downward) deflection at the middle of an unsymmetrical beam is

$$y_{10} = \frac{M_A L^2}{10{,}000\,EI}\left[\frac{0.6}{r_1} + \frac{2.5}{r_2} + \frac{5.6}{r_3} + \frac{10.0}{r_4} + \frac{15.6}{r_5} + \frac{22.5}{r_6} + \frac{30.6}{r_7} + \frac{40.0}{r_8} + \frac{50.6}{r_9} + \frac{60.4}{r_{10}} + \frac{62.0}{r_{11}} + \frac{60.0}{r_{12}} + \frac{56.8}{r_{13}} + \frac{52.5}{r_{14}} + \frac{46.9}{r_{15}} + \frac{40.0}{r_{16}} + \frac{31.9}{r_{17}} + \frac{22.5}{r_{18}} + \frac{11.9}{r_{19}} + \frac{2.1}{r_{20}}\right], \quad (16)$$

where M_A (Fig. 4) = restraining moment at the left end of the beam in question; L = length of span; I_0 = I(minimum); and E = modulus of elasticity.

If the beam is symmetrical, where $r_{20} = r_0$, $r_{19} = r_1$, $r_{18} = r_2$, etc, Eq. (16) becomes

$$y_{10} = \frac{M_A L^2}{10{,}000\, EI_0}\left[\frac{2.1}{r_0} + \frac{12.5}{r_1} + \frac{25.0}{r_2} + \frac{37.5}{r_3} + \frac{50.0}{r_4} + \frac{62.5}{r_5} + \frac{75.0}{r_6} + \frac{87.5}{r_7} + \frac{100.0}{r_8} + \frac{112.5}{r_9} + \frac{60.4}{r_{10}}\right]. \tag{17}$$

If the r's $= 1$ (I constant),

$$y_{10} = \frac{M_A L^2}{10{,}000\, EI}\,[625] = \frac{1}{16}\, M_A L^2,$$

which is a check, since y_{10} (for the unit conjugate beam) $= \frac{1}{6} \times \frac{1}{2} - \frac{1}{2} \times \frac{1}{2} \times \frac{1}{2} \times \frac{1}{3} \times \frac{1}{2} = \frac{1}{16}$.

By an analogous procedure, deflections caused by M_B alone at the right end of beam AB may be found; and the same formulas may be used if the numbering is reckoned from right to left and M_B is substituted for M_A.

MATRICES WITH ALL THEIR CHARACTERISTIC ROOTS IN THE INTERIOR OF THE UNIT CIRCLE

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In economics it is of interest to have criteria that a given square matrix $A = (a_{\kappa\lambda})$ of order n has all of its characteristic roots in the interior of the unit circle. We set

$$R_\lambda = \sum_{\kappa=1}^{n} |a_{\kappa\lambda}| \qquad (\lambda = 1, 2, \cdots, n),$$

$\max R_\lambda = R$, and $\min R_\lambda = r$. Assume that A is unreduced, that is, it is impossible to distribute the numbers $1, 2, \cdots, n$ into two sets $\alpha_1, \alpha_2, \cdots \alpha_k$ and $\beta_1, \beta_2, \cdots, \beta_{n-k}$ such that all the elements vanish which the rows $\alpha_1, \alpha_2, \cdots, \alpha_k$ have in common with the columns $\beta_1, \beta_2, \cdots, \beta_{n-k}$. If $1 \geqq R > r$, then all the characteristic roots of A have an absolute value less than 1 (Solow [6], Brauer [2], [4]).

Solow stated that it would be useful to have similar results for $R > 1$. I shall prove such criteria for the case that only one of the sums R_λ is greater than or equal to 1 or that a power of A has this property.

We need the following lemma.

Lemma. Let a and b be real numbers of absolute value less than 1 and ζ be a point on the boundary of the unit circle. Then

$$|\zeta - a||\zeta - b| \geqq 1 - |a + b| + ab. \tag{1}$$

Proof. If $ab \geqq 0$, then (1) holds since

$$|\zeta - a||\zeta - b| \geqq (|\zeta| - |a|)(|\zeta| - |b|)$$
$$= (1 - |a|)(1 - |b|) = 1 - |a + b| + ab.$$

If $ab < 0$, then we set $\zeta = \xi + i\eta$, whence $\xi^2 + \eta^2 = 1$. Therefore

$$|\zeta - a|^2 = (\xi - a)^2 + \eta^2 = -2a\xi + a^2 + 1,$$
$$|\zeta - b|^2 = (\xi - b)^2 + \eta^2 = -2b\xi + b^2 + 1.$$

Hence $|\zeta - a|^2|\zeta - b|^2$ is a quadratic polynomial in ξ with the leading term $4ab\xi^2$. Therefore the second derivative of this polynomial is negative and the polynomial takes its smallest value in the interval $\{-1 \cdots + 1\}$ at one of the endpoints. It follows that

$$|\zeta - a||\zeta - b| \geqq \min\{(1 - a)(1 - b), (1 + a)(1 + b)\} = 1 - |a + b| + ab.$$

Hence the Lemma is proved.

In my paper [1], I proved the following theorem (Theorem 11):
Let $A = (a_{\kappa\lambda})$ be a square matrix of order n. Set

$$R_\lambda = \sum_{\kappa=1}^{n} |a_{\kappa\lambda}| \qquad (\lambda = 1, 2, \cdots, n). \tag{2}$$

Then each characteristic root lies in the interior or on the boundary of at least one of the $n(n-1)/2$ ovals of Cassini

$$|z - a_{\kappa\kappa}||z - a_{\lambda\lambda}| \leqq (R_\kappa - |a_{\kappa\kappa}|)(R_\lambda - |a_{\lambda\lambda}|) \qquad (\kappa, \lambda = 1, 2, \cdots, n; \kappa \neq \lambda). \tag{3}$$

We use this result to prove the following theorem.

THEOREM 1. Let $A = (a_{\kappa\lambda})$ be a square matrix of order n with real elements and R_λ be defined by (2). Assume that only one of the R_λ, say $R_1 \geqq 1$. If

$$|a_{11}| < 1 \tag{4}$$

and if

$$R_1 - |a_{11}| < \frac{1 - |a_{11} + a_{\lambda\lambda}| + a_{11}a_{\lambda\lambda}}{R_\lambda - |a_{\lambda\lambda}|} \tag{5}$$

for all those λ for which $R_\lambda > |a_{\lambda\lambda}|$, then all the characteristic roots of A lie in the interior of the unit circle.

PROOF. Since

$$R_\lambda < 1 \qquad (\lambda = 2, 3, \cdots, n), \tag{6}$$

we have

$$|a_{\lambda\lambda}| < 1 \qquad (\lambda = 2, 3, \cdots, n). \tag{7}$$

Let ζ be a point on the circumference of the unit circle. Then

$$|\zeta - a_{\kappa\kappa}||\zeta - a_{\lambda\lambda}| \geqq (1 - |a_{\kappa\kappa}|)(1 - |a_{\lambda\lambda}|) \qquad (\kappa, \lambda = 1, 2, \cdots, n; \kappa < \lambda). \tag{8}$$

For $\kappa \neq 1$, it follows from (8) and (6) that

$$|\zeta - a_{\kappa\kappa}||\zeta - a_{\lambda\lambda}| > (R_\kappa - |a_{\kappa\kappa}|)(R_\lambda - |a_{\lambda\lambda}|). \tag{9}$$

If $\kappa = 1$ and $R_\lambda = |a_{\lambda\lambda}|$, then (9) follows from (4) and (7). If finally $\kappa = 1$ and $R_\lambda > |a_{\lambda\lambda}|$, then it follows from the lemma that

$$|\zeta - a_{11}||\zeta - a_{\lambda\lambda}| \geqq 1 - |a_{11} + a_{\lambda\lambda}| + a_{11}a_{\lambda\lambda} \tag{10}$$

and from (5) that

$$1 - |a_{11} + a_{\lambda\lambda}| + a_{11}a_{\lambda\lambda} > (R_1 - |a_{11}|)(R_\lambda - |a_{\lambda\lambda}|). \tag{11}$$

Now (9) is obtained from (10) and (11). Therefore (9) is proved for each case.

Let G be the intersection of the region of the unit circle and the region of one

of the ovals (3). Denote the foci of this oval by F_1 and F_2. Every point P on the boundary of the oval has the property that it can be joined with at least one of the foci, say F_1 by a continuous curve which lies completely in the oval. It follows from (4) and (7) that F_1 and F_2 lie in G, and from (9) that no point of the circumference of the unit circle belongs to G. Hence the curve F_1P cannot intersect the circumference of the unit circle. Thus P must be an interior point of the unit circle. It follows that all the ovals (3), hence all the characteristic roots of A lie in the interior of the unit circle.

This proves Theorem 1.

The following matrix, for instance, satisfies (5)

$$A = \begin{pmatrix} 0 & 1/3 & 1/6 \\ 1/2 & 1/3 & 1/4 \\ 7/12 & 1/4 & 1/2 \end{pmatrix}.$$

Since the characteristic roots of the matrix A^k are the kth powers of the characteristic roots of A, we obtain

THEOREM 2. Let A be a matrix such that the matrix A^k satisfies Theorem 1, then all the characteristic roots of A lie in the interior of the unit circle.

If not all the elements of A have the same sign, then Theorem 1 can be improved. For this purpose we need the following theorem I proved in a paper to be published soon [3].

Set

$$Q_\lambda = R_\lambda - |a_{\lambda\lambda}| \qquad (\lambda = 1, 2, \cdots, n)$$

and

$$(12) \quad Q_{\kappa\lambda} = |a_{\kappa\lambda}| Q_\kappa + |a_{\lambda\kappa}| (Q_\lambda - |a_{\kappa\lambda}|) + \sum_\nu |a_{\nu\kappa} a_{\nu\lambda}| + \sum_{\nu<\mu} |a_{\nu\kappa} a_{\mu\lambda} + a_{\mu\kappa} a_{\nu\lambda}|$$

where κ, λ, μ, and ν run from 1 to n, and where $\kappa \neq \lambda$, $\mu \neq \kappa, \lambda$; $\nu \neq \kappa, \lambda$. Then each characteristic root must lie in the interior or on the boundary of at least one of the $n(n-1)/2$ ovals of Cassini

$$(13) \quad |z - a_{\kappa\kappa}||z - a_{\lambda\lambda}| \leqq Q_{\kappa\lambda} \qquad (\kappa, \lambda = 1, 2, \cdots, n; \kappa \neq \lambda).$$

It follows from (12) that every interior point of one of the ovals (13) is an interior point of at least one of the ovals (3).

Using this result we obtain the following theorem.

THEOREM 3. Let A be a square matrix of order n with real elements $a_{\kappa\lambda}$. Assume that $|a_{\kappa\kappa}| < 1$ for $\kappa = 1, 2, \cdots, n$. Let $Q_{\kappa\lambda}$ be defined by (12). If

$$(14) \quad 1 - |a_{\kappa\kappa} + a_{\lambda\lambda}| + a_{\kappa\kappa}a_{\lambda\lambda} > Q_{\kappa\lambda} \qquad (\kappa, \lambda = 1, 2, \cdots, n; \kappa \neq \lambda),$$

then all the characteristic roots of A lie in the interior of the unit circle.

PROOF. Each characteristic root must lie in the interior or on the boundary of at least one of the ovals (13). But it follows from the Lemma and from (14)

that each point of the circumference of the unit circle lies in the exterior of each of these ovals. The further proof is the same as that of Theorem 2.

G. W. Medlin [5] proved the following theorem.

Let $A = (a_{\kappa\lambda})$ be a square matrix of order n with real elements. Denote the sum of the positive terms in $\sum_{\nu=1}^{n} a_{\nu\kappa}a_{\nu\lambda}$ by $u_{\kappa\lambda}$, the sum of the negative terms by $v_{\kappa\lambda}$, and max $(u_{\kappa\lambda}, |v_{\kappa\lambda}|)$ by $w_{\kappa\lambda}$. We set

$$(15)\quad Q'_{\kappa\lambda} = |a_{\kappa\lambda}|Q_\kappa + |a_{\lambda\kappa}|(Q_\lambda - |a_{\kappa\lambda}|) + w_{\kappa\lambda} + \sum_{\nu<\mu} |a_{\nu\kappa}a_{\mu\lambda}| + \sum_{\nu<\mu} |a_{\nu\lambda}a_{\mu\kappa}|.$$

Then each characteristic root of A must lie in the interior or on the boundary of at least one of the $n(n-1)/2$ ovals

$$|z - a_{\kappa\kappa}||z - a_{\lambda\lambda}| \leqq Q'_{\kappa\lambda}$$

$$(\kappa, \lambda = 1, 2, \cdots, n; \kappa \neq \lambda).$$

Using this result we obtain the following theorem.

THEOREM 4. Theorem 4 remains correct if in (14) $Q_{\kappa\lambda}$ is replaced by $Q'_{\kappa\lambda}$ defined by (15).

In some cases Theorem 3 gives a better result than Theorem 4, in other cases Theorem 4 is better.

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OBSERVATIONS ON THE CILIATE *BURSARIA OVATA*, N. SP.

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Although a number of species of protozoa have been described under the genus name *Bursaria*, all except one have proved to belong to other genera. The one valid species which is recognized by such authorities on taxonomy as Kahl (1932), Calkins (1933), Bhatia (1936), and Kudo (1946) is the large heterotrichous ciliate *B. truncatella* O. F. Müller, 1786. Its structure, division, conjugation, encystment and excystment have been studied by a number of investigators, among them, Brauer (1886), Schuberg (1887), Lund (1917), Schmähl (1926), Poljansky (1934), and Beers (1948). Kahl's brief account (p. 476) of its structure and general biology is excellent. Schmähl's contribution merits special mention, since it is more comprehensive than its title indicates. It includes a study of the structure of the adult, trophic individual, based in part on the reconstruction of a model from serial transverse sections. Thus Schmähl was able to clarify several controversial points, such as the nature of the "Mundspalte" or oral cleft and the structure of the membranelles. It is evident that our knowledge of the morphology of *B. truncatella* is relatively complete.

Since only one species of *Bursaria* is known to date, in spite of much intensive taxonomic work on ciliates, I am somewhat hesitant to describe a new species. Nevertheless, there appeared in September 1946 in a large culture dish in the laboratory considerable numbers of bursarias which did not agree with the descriptions of *B. truncatella* regarding body shape, configuration of the peristomial cavity, food habits, and, most notable of all, the structure of the cyst membranes. This dish contained the water, detritus, and miscellaneous organisms of a recent collection from a temporary pool in Sparrow's Pasture, just north of Chapel Hill. Some hay infusion was added when the material was brought in, and the bursarias flourished for about 3 weeks. In this period various observations concerning structure and food habits were made on active specimens, and several dozen individuals were fixed in Schaudinn's fluid (with acetic) and stained by the Feulgen method. A few specimens were similarly fixed, embedded in paraffin, sectioned transversely at 5μ, and stained in iron hematoxylin. In this process they were handled singly in pipettes and stained provisionally in alcoholic Fast Green to facilitate orientation during embedding. Other individuals when deprived of food in spring water formed resting cysts. The various notes were laid aside in a folder, since I was undecided as to their significance.

Then, in the early part of 1950 one of our graduate students, E. Eugene Jones, Jr., remarked that large numbers of bursarias had developed in a jar of water (rich in organic matter) which he had taken from a small marshy area on the Mann's Chapel Road some 5 miles south of Chapel Hill, and that the specimens were plump, ovoid, and noticeably unlike *B. truncatella*. It was evident that his specimens were quite like those taken by me in 1946. Thus I became convinced

that there are in the vicinity of Chapel Hill two valid species of *Bursaria*, *viz.*, the well-known, cosmopolitan *B. truncatella* and a second which I shall designate as *B. ovata*, n. sp.

Bursaria ovata, n. sp.

Length, 500–1000μ; width, 250–475μ; dorso-ventral thickness, 150–240μ. Body elongate-ovate, twice as long as wide; convex dorsally, flattened ventrally, obliquely truncate anteriorly, narrowed and tapering posteriorly, the bluntly pointed posterior end turned slightly toward the left; greatest width in the middle third, greatest dorso-ventral thickness between first and middle thirds; the margin of the longer right side convex, that of the shorter left side only very slightly convex or nearly straight. Anterior peristomial field and ventral peristomial cleft relatively narrow. Terminal, tubular portion of peristomial cavity long and U-shaped, narrowing gradually toward the mouth (cytostome), the tube itself extending posteriorly on the right side of the body, then bending to the left, and finally continuing anteriorly on the left side to end in the mouth, which is in the anterior half of the body. Peristomial groove restricted to right margin of peristome. Stomatoplasm beginning posterior to the ventral peristomial cleft and hence embracing only the posterior end of the groove, then continuing almost to the mouth as a circular cord lying dorsal to the peristomial tube. Food chiefly bacterial zooglea and small flagellates, supplemented by *Arcella* and some small ciliates such as *Euplotes*; food vacuoles very numerous. Resting cyst spherical, 230–280μ in diameter, with two membranes and an emergence pore. The ectocyst very sticky and much folded, its external surface presenting an intricate and irregular pattern of ridges and depressions; some depressions shallow, others deep enough to permit contact with the endocyst, but bridges uniting ectocyst and endocyst absent. Ciliation, nuclei, and contractile vacuoles as in *B. truncatella*.

Figure 1 shows the general shape of the body of *B. ovata*, as well as the proportions of the anterior peristomial field (AP) and the ventral peristomial cleft (PC), both of which are relatively wider in *B. truncatella*. It is evident that the shape of *B. ovata* differs significantly from that of *B. truncatella*, whose body form is described by Bhatia (p. 241) as "broadly ovate, purse- or sac-shaped, ... scarcely one and a half times as long as broad, ... widest posteriorly . . . ." It may be added that in *B. truncatella* the greatest dorso-ventral thickness occurs in the posterior third of the body (Kahl, p. 476).

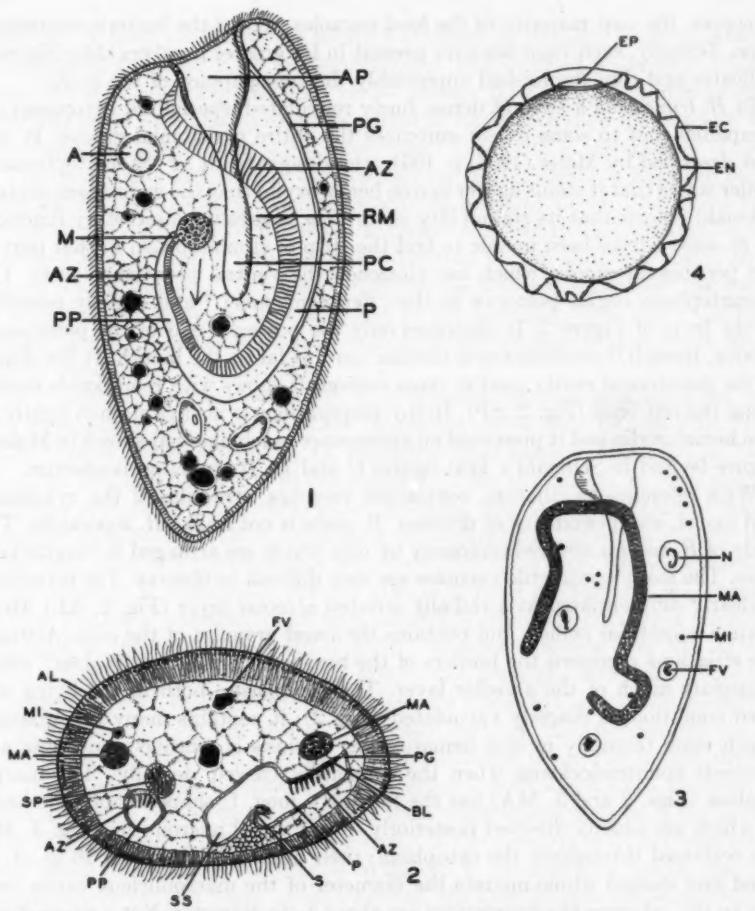
The remarkably long peristomial tube of *B. ovata* is worthy of special comment. The adoral membranelles begin as a narrow zone on the left side of the anterior peristomial field (Fig. 1, AZ). The zone proceeds dorsally and to the right, the membranelles widening as they pass across the dorsal wall of the peristomial cleft, though from this point they decrease gradually in width as the mouth is approached. The adoral zone now continues approximately parallel to the ciliated right margin or lip (RM) of the ventral cleft. This margin, as in *B. truncatella*, bears a ciliated ridge or septum which projects dorsally, though the septum can rarely be seen in living specimens and is not shown in Figure 1. At the level of the posterior margin or angle of the ventral peristomial cleft ("Peristomwinkel" of Schmähl), the peristome takes the form of a closed tube (called gullet by Lund, 1917) or peristomial cavity (Figs. 1 and 2, P) into which the septum (Fig. 2, S)

projects and thus marks off a non-ciliated so-called septal space (SS). Somewhat posterior to the level of the "Peristomwinkel," the septum disappears, the adoral zone turns toward the left, and the peristomial cavity narrows (Fig. 1). Well toward the posterior end of the animal, the adoral zone and accompanying peristomial cavity bend forward and proceed anteriorly. The peristomial cavity continues to narrow (Figs. 1 and 2, PP), until it finally reaches the mouth (Fig. 1, M), which actually lies in the anterior half of the animal. Thus the peristomial cavity or tube is U-shaped and much longer than in *B. truncatella*. For descriptive purposes it may be said to consist of a wide right limb, a posterior curvature, and a narrow left limb. The formation of food vacuoles may be readily observed in individuals that are creeping about slowly in a watch glass and feeding at the bottom. As each vacuole becomes detached from the end of the peristomial tube, cyclosis carries it posteriorly in the direction of the arrow in Figure 1.

As in *B. truncatella*, each adoral membranelle rests on a basal lamella (Fig. 2, BL). Because of the relative thickness of the sections and the curvature of the peristomial cavity, several membranelles and their respective lamellae appear in each limb in Figure 2. The right limb (P) is undoubtedly somewhat narrowed dorso-ventrally as a result of the microtechnical procedure, though the left limb (PP) appears to have retained its normal shape. Except in the areas of the membraneiles and septum, the peristomial tube is non-ciliated.

Dorso-laterally the right margin of the peristomial area presents a peristomial groove (Figs. 1 and 2, PG) which in general is coextensive with the septum. The function of a similar though relatively longer groove in *B. truncatella*, designated by Brauer as the "Peristomrinne," by Schuberg as the "Mundspalte," has been the subject of much discussion, but the experimental evidence indicates that it is not the functional mouth. For example, Lund (1914, p. 6) states that "there is but one path of acceptance for both large and small particles," namely, by way of the inner, terminal opening of the peristomial tube. He states further (p. 7) that the ingestion of large particles is facilitated by a peristaltic wave along its wall. With reference to the ingestion of the large ciliates *Stentor* and *Frontonia*, Schmähl (p. 379) writes as follows: "Stets erfolgte eine Überführung der Nahrungstiere in das Entoplasma an der Mündung der Peristomhöhle." The formation of food vacuoles at the inner terminus of the peristomial tube of *B. ovata* has already been mentioned.

Apparently *B. ovata* regularly utilizes bacteria as food, whereas *B. truncatella* rarely feeds on them (Lund, 1914, p. 3). I have watched repeatedly in *B. ovata* the formation of food vacuoles which appeared to contain nothing but bacteria in the form of zooglea, or a combination of bacteria, small flagellates, and probably detritus, even though the medium contained many kinds of ciliates, as well as an abundance of the shelled rhizopod *Arcella*. However, *Arcella* was sometimes ingested (Fig. 1, A), as was the hypotrich *Euplotes*. Unfortunately, I was never able to observe the actual ingestion of an arcella in order to see whether it entered by the true mouth or by the "Mundspalte." In Feulgen preparations some of the food vacuoles occasionally contained Feulgen-positive material, undoubtedly the nuclei of ingested ciliates. The slender C-shaped bodies in some vacuoles were clearly the nuclei of ingested individuals of *Euplotes* (Fig. 3, N).



FIGURES 1–4. The heterotrichous ciliate *Bursaria ovata*, n. sp.

FIG. 1. A large active individual in dorsal view, sketched from life with the aid of the camera lucida. × 100.

FIG. 2. Cross section of active individual just posterior to ventral peristomial cleft (at level of label line P of Fig. 1). Schaudinn-iron hematoxylin. Camera lucida. × 150.

FIG. 3. Whole mount of a medium-sized active specimen to show nuclei. Schaudinn-Feulgen. Camera lucida. × 100.

FIG. 4. Resting cyst in optical section. Drawn from life with the aid of the camera lucida. × 140.

A, ingested *Arcella;* AL, alveolar layer; AP, anterior peristomial field; AZ, adoral zone of membranelles; BL, basal lamella; EC, ectocyst; EN, endocyst; EP, emergence pore; FV, food vacuole; M, mouth (cytostome); MA, macronucleus; MI, micronucleus; N, nucleus of ingested *Euplotes;* P, peristomial cavity, wide right limb; PC, ventral peristomial cleft; PG, peristomial groove; PP, peristomial cavity, narrow left limb; RM, right margin of peristomial cleft; S, septum; SP, stomatoplasm; SS, septal space.

However, the vast majority of the food vacuoles were of the bacteria-containing type. Actually, such vacuoles were present in far greater numbers than Figure 1 indicates and thus diminished appreciably the transparency of the body.

In *B. truncatella* a strip of dense, finely reticulated, specialized cytoplasm accompanies and to some extent embraces the entire peristomial groove. It was first described by Maier (1903, p. 105), who designated it as "Stomatoplasma." Maier states that it stains lighter in iron hematoxylin than the endoplasm, though Schmähl reports that its stainability is variable, suggesting a secretory function. In *B. ovata* I have been unable to find the strip of stomatoplasm in that part of the peristomial groove which lies alongside the ventral peristomial cleft. The stomatoplasm begins posterior to the "Peristomwinkel," and slightly posterior to the level of Figure 2. It embraces only the posterior end of the peristomial groove, though it continues as a circular cord as far as the mouth. It lies dorsal to the peristomial cavity, and in cross sections it shows with remarkable clarity along the left limb (Fig. 2, SP). In my preparations it always stained lightly in iron hematoxylin and it presented an appearance much like that shown in Maier's Figure 9e and in Schmähl's Text-figures C and D. Its function is obscure.

With reference to ciliation, contractile vacuoles, structure of the cytoplasm and nuclei, and periodicity of division, *B. ovata* is not unlike *B. truncatella*. The body of *B. ovata* is covered uniformly by cilia which are arranged in longitudinal rows. The many contractile vacuoles are very difficult to observe. The ectoplasm is clearly differentiated as a radially striated alveolar layer (Fig. 2, AL) which bears a superficial pellicle and contains the basal granules of the cilia. Actually the striations represent the borders of the hexagonal prisms or "Waben" which constitute much of the alveolar layer. The endoplasm, both in the living and fixed condition, is coarsely vacuolated (Fig. 2); it contains many fine granules which stain intensely in iron hematoxylin. Cyclosis is relatively energetic and proceeds counterclockwise when the animal is viewed dorsally. The macronucleus (Figs. 2 and 3, MA) has the form of a long, U-shaped band, the limbs of which are usually directed posteriorly. Many small micronuclei (Fig. 3, MI) are scattered throughout the cytoplasm; their number varies from 16 to 24. In fixed and stained whole mounts the diameter of the macronucleus varies from 12μ to 16μ, whereas the micronuclei are about 4μ in diameter. Not a single dividing specimen was seen in the course of the usual work day, yet the number of individuals increased greatly in the original culture dish. Thus it is evident that *B. ovata*, like *B. truncatella*, exhibits the peculiar and unexplained habit of nocturnal division.

The formation of cysts in the absence of food has been mentioned. In order to obtain cysts, active specimens of *B. ovata* were transferred to 3-cc. amounts of spring water (pH 6.8) contained in covered Boveri dishes. In these, encystment invariably occurred within 24 hours. Medium-sized indiviuals formed one cyst each; some very large individuals divided once and thus produced two cysts. The original number of individuals per Boveri dish, whether 10, 50, or 100, had no effect on the encystment process. Thus crowding was not a factor in the production of cysts, and the evidence indicates that encystment resulted solely

from lack of food. Each cyst was securely attached by its outer membrane to the inner wall of the dish. With the addition of a little distilled water from time to time, the cysts were kept without visible change for 8 months. Unfortunately, time was never available during this period to undertake their excystment, and they finally perished during my absence from Chapel Hill in the summer of 1947.

The remarkable structure of the resting cyst of *B. truncatella* has been recognized for nearly a century. (Reference may be made to Beers, 1948, for a brief review of the literature.) The cyst is spherical and has two membranes, a thin endocyst and a thicker ectocyst whose relation to the endocyst seems to be unique among ciliates. The outer surface of the ectocyst is faceted; the center of each facet is depressed and attached to the endocyst by a low, cylindrical bridge. One of the facets bears at its center a closed emergence pore which superficially resembles a large bridge.

In *B. ovata* the resting cyst (Fig. 4) is likewise spherical and provided with two membranes. The diameter of the protoplast varies from 190μ to 240μ (215μ in the typical cyst shown in Fig. 4). The membranes measure about 20μ in thickness and thus add about 40μ to the diameter of the protoplast. In close contact with the protoplast is the thin, inner endocyst (Fig. 4, EN). External to the endocyst is the ectocyst (EC), the surface of which is folded in such a manner that ridges and intervening depressions occur over the entire cyst. These depressions, unlike those of *B. truncatella*, are not always circular or regularly polygonal in outline, but are of variable shape, size, and depth. Some are circular or elliptical, but most of them take the form of short, irregular channels over the surface. Some depressions are relatively deep, and in such areas the ectocyst is in contact with the endocyst, though other depressions are shallow (Fig. 4). Bridges uniting ectocyst and endocyst are absent. The space between the two membranes appears to be occupied by a homogeneous, transparent fluid. The largest depression in the ectocyst is occupied by the emergence pore which has a diameter of about 40μ. It is closed by an opercular membrane which rests on a low collar, as in *B. truncatella*. A noteworthy property of the ectocyst is its extreme stickiness, which makes the cysts difficult to handle. If touched by a needle, the cyst adheres firmly to it; if taken up in a pipette, the cyst usually lodges securely in the lumen. Thus the membranes, both in structure and physical properties, differ significantly from those of *B. truncatella*.

SUMMARY

The heterotrichous ciliate *Bursaria ovata*, n. sp., is described. Features which distinguish it from *B. truncatella* are the following: The elongate-ovate body, which tapers posteriorly and bends slightly to the left; the long, U-shaped peristomial cavity; certain characters of the cyst; and the utilization of bacteria, chiefly zooglea, as the principal article of food.

In the absence of food *B. ovata* readily forms spherical resting cysts which have two membranes and an emergence pore. The outer surface of the ectocyst is very sticky and much folded. The cyst lacks the peculiar bridges which connect ectocyst and endocyst in *B. truncatella*.

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NEW RECORDS OF RHEOPHILIC WATER-MITES FROM NORTHWESTERN NORTH CAROLINA

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Plates 16 and 17

Seven species of water-mites, all of which represent first records in the United States, are herein reported from material transmitted to the writer by Dr. Joseph R. Bailey of Duke University. This material was recovered from collections taken during the summer of 1949 in connection with a stream survey program sponsored by the North Carolina Wildlife Resources Commission.

All specimens were taken in the six northwesternmost counties of North Carolina, near the Virginia and Tennessee borders of the state. (Approximate range 81° to 82°21′ W by 35°55′ to 36°30′ N.)

Five families of Hydracarina are represented in the collections. Among the identified material, two families are represented by two species each, the others by one species each. Two nymphs included among the water-mites found were not identified.

Previously known distributional notes are given for each of the species. References which will aid in the identification of these forms are included. No attempt has been made to make the citations exhaustive. Keys to the families will be found in Baker and Wharton (1952).

Super-Family Lebertiae

Sperchonidae:

Sperchon (*sensu strictu*) *squamosus* Kramer, 1879.
(Plate 16: Figs. 1–2)

2♂ Avery Co., N. Toe River, 1 mi. s.e. of Minneapolis, 25 Aug., '49;
2♀ Avery Co., N. Toe River, 1 mi. n. of Ingalls, 25 Aug., '49;
2♀ Alleghany Co., Little River, 1.5 mi. e. of Sparta, 16 June, '49;
4♂ Wilkes Co., Roaring River, 1.5 mi. below junction of 3 prongs, 25 July, '49;
2♀ Watauga Co., South fork of New River, Todd, 8 Aug., '49.

This species has a weak, leathery skin. The color is brownish-red, with a paler marginal zone. Two small, bean-shaped sclerites are present in the mid-dorsal region. External genitalia consists of two movable plates with three pairs of genital suckers. A pre-genital sclerite is present in males, absent in females.

Previously reported from: Bohemia, Belgium, France, Germany, Gt. Britain, Italy, Norway, Russia, Switzerland (Soar and Williamson, Vol. I, p. 98); Bornholm (Lundblad, 1930, p. 8); Germany (Viets, 1936, p. 152); Germany, Norway (Piersig, 1901, p. 167); Russia (Sokolow, 1940, p. 173).

Sperchon (*s.s.*) *longirostris* Koenike, 1895.

(Plate 16: Figs. 3–4)

1♂ Watauga Co., South Fork of New River, Todd, 8 Aug., '49.

This form is very much like *S. squamosus*, and in fact its specific identity has been questioned (Viets, 1936, p. 153). Differences in palpal proportions and setal distributions seem to warrant separation. *S. longirostris* also has elaborate claw plates which are absent in *S. squamosus*.

Previously reported from: France, Germany, Ireland, Italy, Switzerland (Viets, 1936, p. 153); Ireland, Italy, Sweden, Switzerland (Soar and Williamson, Vol. I, p. 105); Rhatikon, Gorschina-See (Piersig, 1901, p. 167).

LEBERTIIDAE:

Lebertia (*s. s.*) *natans* Viets, 1926.

(Plate 16: Figs. 5–7)

4 (sex?) Yancey Co., Cane River, 5.2 mi. w. of Burnsville, 2 Sept., '49;

1 (sex?) Yancey Co., Cane River, 3.5 mi. s.w. of Burnsville, 7 Sept., '49.

This species exhibits extensive ventral sclerotization, reaching nearly to the posterior margin; a broad indentation in the posterior epimeral margin forms the genital field. The genital plates have three pairs of genital suckers. The dorsum exhibits sparse sclerotization, as shown in Figure 6.

Previously reported from: Germany (Viets, 1926, p. 65; Viets, 1936, p. 185).

In addition to the above species there are nine adult and one nymphal Lebertiids which fall into two groups, representing two undetermined species of the genus *Lebertia* (*sensu strictu*).

ATRACTIDEIDAE:

Atractides (*sensu strictu*) *anomalous* C. L. Koch, 1837.

(Plate 16: Figs. 8–11)

1♀ Alleghany Co., Little River, 1.5 mi. e. of Sparta, 16 June, '49;

1♀ Wilkes Co., Reddies River, 4 mi. e. of Mulberry, 26 July, '49.

This species is widely distributed, though not usually occurring in large numbers; it is generally found in slow-flowing waters. The ventral epimera join behind the genital area, completely enclosing it. The dorsum is completely sclerotized, there being two pairs of elongated anterior plates, with a larger plate covering most of the dorsal surface.

Previously reported from; Algeria, Austria, Germany, Gt. Britain, Holland (?), Hungary, Italy, Russia, Switzerland, Turkestan (Viets, 1936, p. 235); Germany, Italy, Russia, Switzerland, Turkestan (Soar and Williamson, Vol. II, p. 89); Germany (Piersig, 1901, p. 138).

SUPER-FAMILY PIONAE

HYGROBATIDAE:

Hygrobates (*sensu strictu*) *fluviatilis* (Ström 1768).

(Plate 17: Figs. 12–13)

1♀ Yancey Co., Cane River, 5.2 mi. w. of Burnsville, 2 Sept., '49.

Mites of this species have a weak-skinned body, yellowish in color and flecked

with brown. The palpal femur bears a toothed peg; the palpal genu is toothed distally on the flexor surface. The anterior epimera are completely fused medially. The suture between epimeral plates III and IV is incomplete medially.

Previously reported from: Holland (Besseling, 1942, p. 2); England, Germany, Switzerland (Piersig, 1901, p. 187); Bohemia, Denmark, France, Germany, Norway, Russia, Scotland, Switzerland (Soar and Williamson, Vol. II, p. 105); Russia (Sokolow, 1940, p. 287); Europe (Viets, 1936, p. 254).

Megapus (*sensu strictu*) *walteri* Viets, 1925.

(Plate 17: Figs. 14–15)

1 ♀ Ashe Co., North Fork of New River, 0.8 mi. n.e. of Creston, 11 July, '49.

This is a thick-skinned species, with distinct plates bearing glandular openings in the extra-epimeral areas. Posterior medial projections of the anterior epimera of this species reach a point nearly opposite the posterior margins of the fourth epimera. A prominent suture is apparent between the anterior epimera.

Previously reported from: France (Viets, 1925, p. 536; Viets, 1936, p. 276).

SUPER-FAMILY AXONOPSAE

AXONOPSIDAE:

Aturus intermedius Protz, 1900

(Plate 17; Figs. 16–19)

1 ♂ Alleghany Co., Little River, 4 mi. s. of Whitehead, 23 June, '49.

This is the smallest of the mites taken in the present collections. The posterior medial edge is marked by a deep incision flanked by elongated, clavate projections. Numerous fine setae arise from the posterior margin of the body. Leg IV, segment 4, bears prominent saber-shaped bristles as indicated in Figure 19.

Previously reported from: Germany (Piersig, 1901, p. 135); Germany, Holland, Ireland, Switzerland (Soar and Williamson, Vol. III, p. 51); Russia (Sokolow, 1940, p. 410); Crimea, East Prussia, France, Holland, Ireland, Rumania, Switzerland (Viets, 1936, p. 382).

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EXPLANATION OF PLATES

(SCALE: Dorsal and ventral shields, Figs. 1, 3, 5, 6, 8, 9, 12, 14, 16, 17; scale represents 0.5 mm. Dissected parts, Figs. 2, 4, 7, 10, 11, 13, 15, 18, 19; scale represents 0.25 mm.)

PLATE 16

Fig. 1. *Sperchon squamosus*, venter ♂.
Fig. 2. *S. squamosus*, medial view, right palp ♂.
Fig. 3. *S. longirostris*, venter ♂.
Fig. 4. *S. longirostris*, lateral view, left palp ♂.
Fig. 5. *Lebertia natans*, venter.
Fig. 6. *L. natans*, dorsum.
Fig. 7. *L. natans*, medial view, left palp.
Fig. 8. *Atractides anomalous*, venter ♀.
Fig. 9. *A. anomalous*, dorsum ♀.
Fig. 10. *A. anomalous*, medial view, left chelicera.
Fig. 11. *A. anomalous*, medial view, left palp.

PLATE 17

Fig. 12. *Hygrobates fluviatilis*, venter ♀.
Fig. 13. *H. fluviatilis*, medial view, right palp.
Fig. 14. *Megapus walteri*, venter ♀.
Fig. 15. *M. walteri*, medial view, left palp ♀.
Fig. 16. *Aturus intermedius*, venter.
Fig. 17. *A. intermedius*, dorsum.
Fig. 18. *A. intermedius*, lateral view, right palp.
Fig. 19. *A. intermedius*, right leg IV, segment 4.

PLATE 16

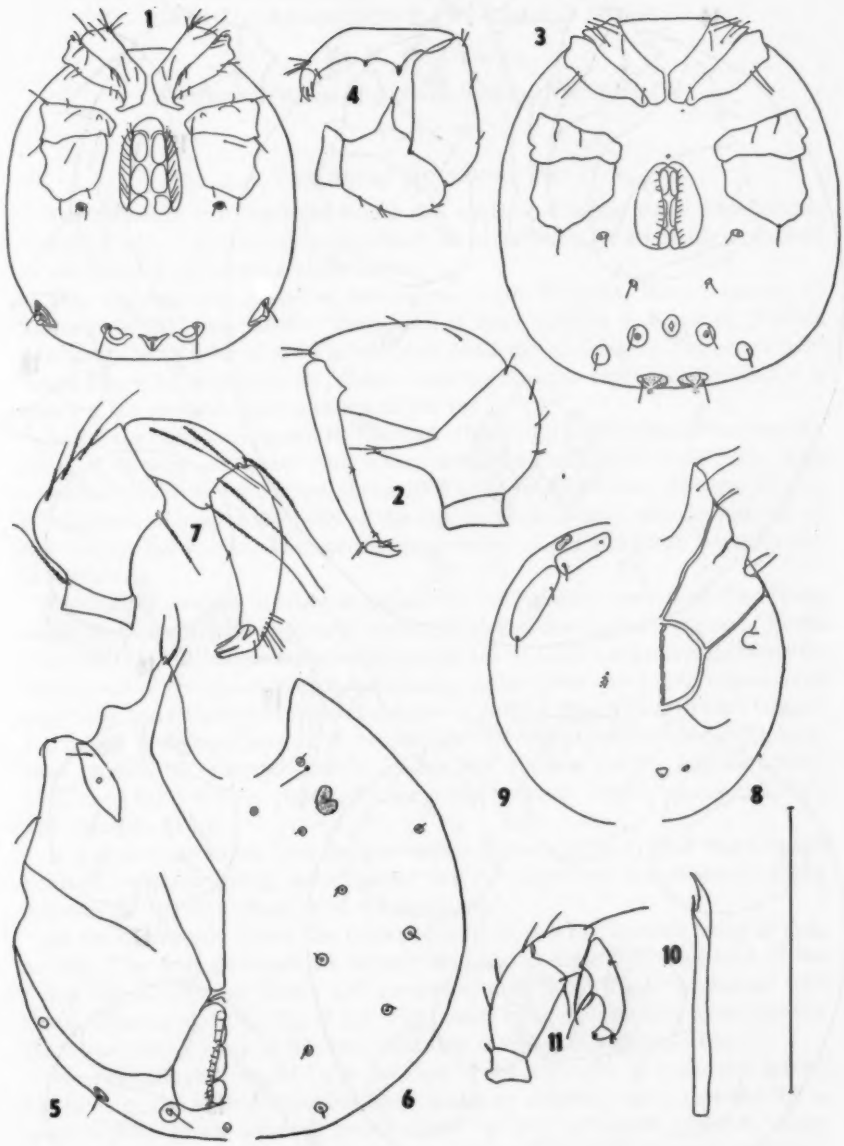
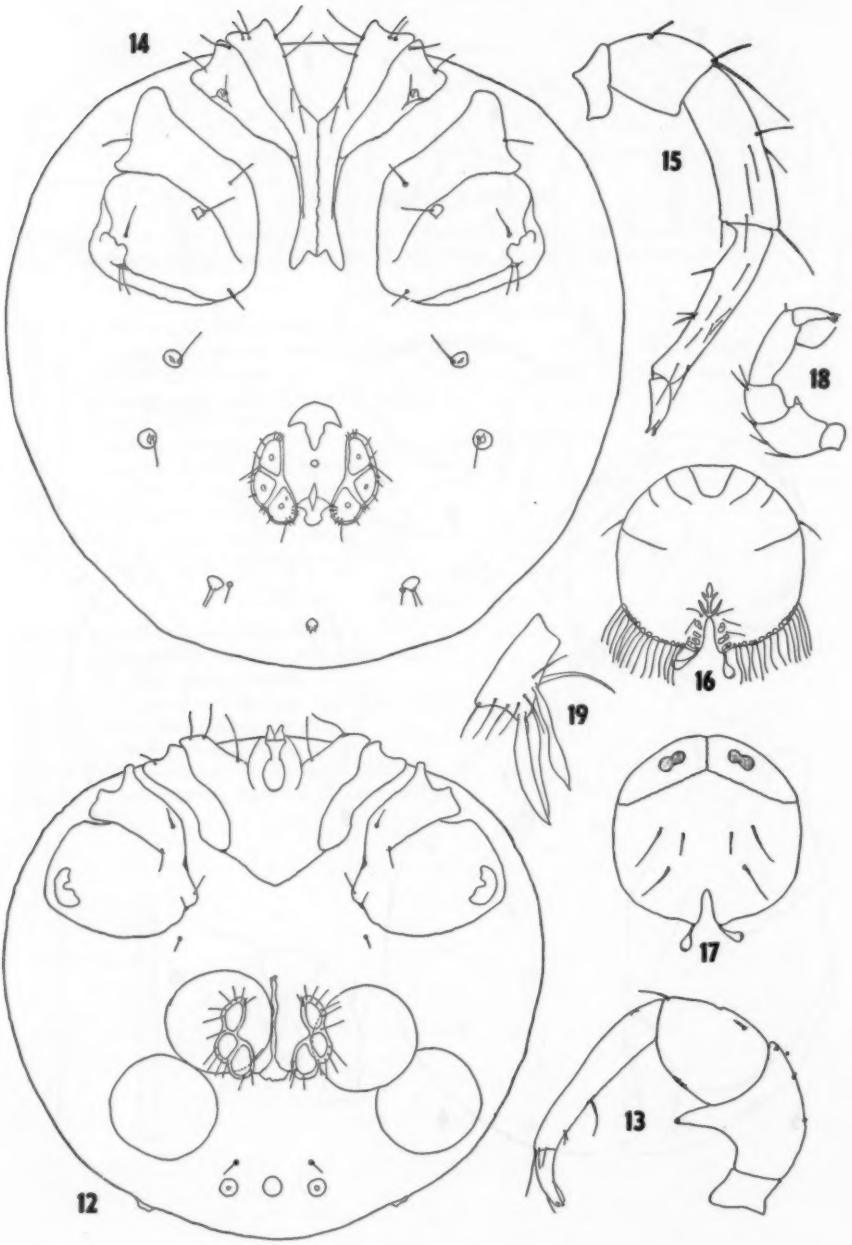


PLATE 17



THE BUTTERFLY RAY, *PTEROPLATEA MACLURA*, AND ITS INTRA-UTERINE-FIN-TAILED EMBRYO

By E. W. Gudger

American Museum of Natural History, New York City

Plate 18

The Adult Butterfly Ray

The butterfly ray, recorded along the western Atlantic coast from Rhode Island, U. S. A., to Surinam in northern South America, is especially abundant in the Beaufort, North Carolina, region.

This ray was first described and figured from Newport, Rhode Island, by LeSueur in 1817 (pp. 41–42). He named it *Raja maclura* in honor of William Maclure, the founder of the Philadelphia Academy of Sciences. LeSueur's published figure is, unfortunately, faded—too much so for reproduction—but it is surely a fair general representation of our ray.

Later, the ray was renamed by Garman (1907, p. 414) *Pteroplatea micrura* because of its very short tail—only about one-third the length of the body. This name had however been given about 1888 to an Indian Ocean *Pteroplatea* with a tail about as long as the body of the fish, but still notably the shortest tail on any Indian ray known. The specific name today of our American butterfly ray is *maclura*.

Pteroplatea maclura is fairly abundant on our Atlantic coast from New York south, but is especially abundant in the Beaufort-Cape Lookout region of North Carolina, U. S. A., where my specimens were taken. Cape Lookout and the southwest trend of the coast from it to Beaufort Inlet make this whole region—and especially the Beaufort-Morehead waters—a natural trap for northward migrating fishes. The abundance of *P. maclura* in this region is attested by the long-time records of: Yarrow (1877); Jordan and Gilbert (1879); Jordan (1880); Jenkins (1887); Wilson (1900); Linton (1905); Smith (1907); Gudger (1911); and Radcliffe (1916).

It is a privilege to use here the fine outline drawing (Fig. 1) from Bigelow and Schroeder's forthcoming encyclopedic work, "Fishes of the Western North Atlantic II: Skates, Rays, and Chimaeroids."

As the illustration shows, the flattened body of this ray is about twice as wide as long. The wide pectorals are bluntly rounded at their tips. The color of the living ray is brownish above and somewhat marbled in fresh specimens. The shape, coloring and flapping of the wide wings in swimming have given the fish the name butterfly ray; it has been said that it "flies through the water."

Some rays have a caudal fin at the base of the tail (with or without a spine), but none of the authorities consulted, figures or describes such a caudal fin or spine in *Pteroplatea maclura*. Smith (1907, p. 45), a careful observer, says, however, that it has no tail fin and that a spine is "usually [always?] lacking." It is known that old and large specimens sometimes have a spine.

The tail is very short—only about one-third the length of the body, as Fig. 1 shows. It is the shortest tail of any ray known to the writer. In keeping with the length of the tail, the pelvic fins are also very small. This insignificant tail has a certain structure of some interest to us.

It was early noted that small ridges are present on the dorsal and ventral surfaces of the tail. Their origin has not hitherto been explained. Garman (1913, p. 414) says: "Tail slender tapering with a distinct fold above and a weaker one below." These folds or ridges were noted in my specimens (Gudger, 1911) as "very faint traces of dorsal and ventral finfolds." W. C. Schroeder has very kindly

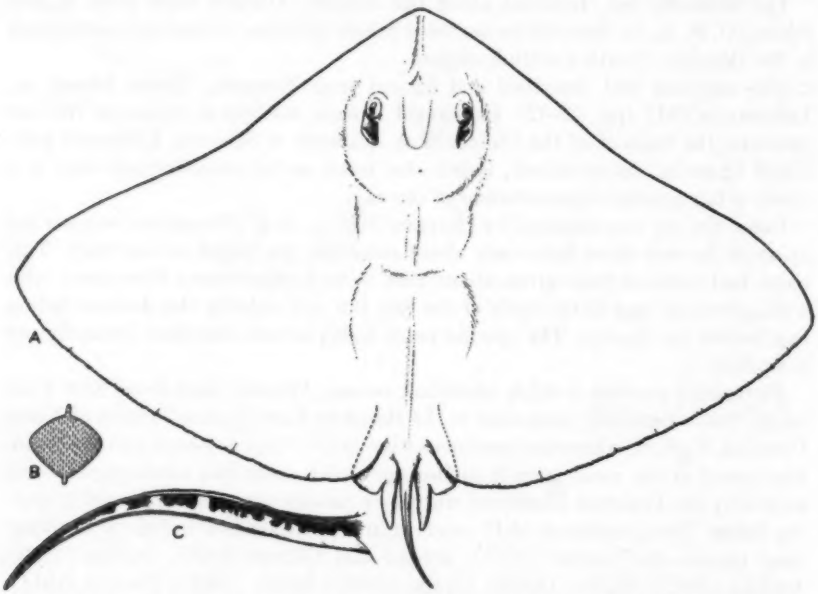


FIG. 1. An adult male *Pteroplatea maclura*. Note the very short tail with the barely perceptible ridge on top, and the small pelvic fins and claspers. B, cross-section of the tail showing dorsal and ventral ridges. C, a drawing showing the four blotches on the dorsal surface of the tail.

Reproduced, with reduction, by courtesy of Bigelow and Schroeder, 1952.

examined two adult specimens of *P. maclura* in the collection of the Museum of Comparative Zoology in Cambridge, Massachusetts, and writes that "*P. maclura* has a ridge on the tail above and below. This ridge is not prominent, but is easily seen." These ridges are shown for the first time in Bigelow and Schroeder's figure of the cross section of the tail of *Pteroplatea maclura* (Fig. 1, B, herein).

Smith (1907) says of the adult ". . . tail with four dark splotches (on upper surface)." Garman (1913) figures (Pl. 33, fig. 3) four dark splotches, plus a fifth at the base of the tail. Bigelow and Schroeder give a detail sketch of the tail (Fig. 1, C, herein), showing an anterior splotch covering nearly half the length

of the tail, two intermediate short blotches, and an elongated posterior one. Being intensely interested in the intra-uterine young, I unfortunately did not note these caudal splothces in my adult specimen; nor can I suggest their origin.

THE FIN-TAILED INTRA-UTERINE EMBRYO

The embryos shown in Plate 18 were taken from the uterine enlargement of the oviduct of the mother, May 27, 1910, as noted by the writer in 1911 (p. 943), 1912 (pp. 148–160), and 1913 (pp. 100–101). The eggshells were gone and the embryos were free in the lumen of the maternal uterus. Three eggs and embryos were photographed. Two appear in dorsal and one in ventral aspect.

The yolksacs are much flattened and are highly vascularized, showing that much of the nutritive yolk has been used up in the growth of the embryos, which are well developed.

One's attention is at once attracted by the great profusion and length of the external gill filaments, emerging from the ventral gill slits. What function do they serve?

The rays are strictly ovo-viviparous—their whole prenatal development takes place as the young lie in the maternal uterus. Some shark embryos form placentas out of their highly vascularized empty yolksacs which become interdigitated with similar folds of the uterine mucosa to form yolksac placentas. But these rays never form such placentas. The young lie free in the maternal uterus in a milk-like fluid supplied by the hair-like villi or trophonemata developed from the inner wall of the uterus. Seemingly the only function these profuse "gill filaments" can have is the absorption of the nutriment. Fed thus, the embryos grow rapidly. In the later stages of the young *Pteroplatea micrura* of Indian seas, the young rays suck into their spiracles clusters of the uterine villi, as figured and described previously (Gudger, 1951, p. 81). Radcliffe (1916, p. 277) describes an identical state of affairs in a *P. maclura* taken at Beaufort, North Carolina. Thus the villi excrete their milky fluid directly into the pharynx and stomach of the young ray. Force-fed, the embryo grows rapidly, and, when extruded, is of relatively large size (about one-third the width of the mother); at once it opens its rolled-up wings and swims away.

The embryos of *Pteroplatea maclura* shown in the photograph (Plate 18) are well developed. The pectoral fins are seemingly united with the head. Pelvic fins are small, as they are in the adult, but are especially noticeable in the little ray (lower left) lying on its back.

Most notable of all the embryonic structures is the tail, about two-thirds as long as the body of the embryo, not the relatively pygmy appendage found in the adult (Fig. 1). Furthermore, each tail bears a thin diaphanous fin above and below. This is best shown in the lower left raylet. The apparent white bands of some supporting tissue in the tail fins of embryos at right and at upper left are merely folds in the diaphanous tissue of the fins.

As the embryos grow older and larger, these fins grow smaller (presumably by absorption). The older intra-uterine embryos were totally devoid of fins. This double fin is truly an embryonic structure.

It seems that the ridges, found on the dorsal and ventral median surfaces of

the tail of the adult *Pteroplatea maclura* (Fig. 1, B), are the remnants of the bases of these embryonic fins. One can only conjecture that they are embryonic reminders of fins possessed by some (selachian?) ancestor.

These fins, described quite incidentally by the writer in 1911, 1912, and 1913, but never before figured, are of special interest as embryonic structures lacking not only in the adults, but even in older intra-uterine embryos, as my notes record. There is no trace of them in the young approaching parturition.

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PLATE 18

Intra-uterine embryos of *Pteroplatea maclura*. The lower left is in ventral aspect, the others are seen from above. Note the flattened yolksacs and the profusion of long external gill filaments emerging from the ventral gill slits. Of most interest are the fins on each tail. These tails are somewhat twisted so that the fins lie flat.

LIFE HISTORY AND ECOLOGIC OBSERVATIONS ON THE TUNDRA MOUSE (*MICROTUS OECONOMUS*) AND LEMMINGS (*LEMMUS TRIMUCRONATUS* AND *DICROSTONYX GROENLANDICUS*) AT BARTER ISLAND, ALASKA

By Frederick S. Barkalow, Jr.

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Plate 19

The writer was privileged to spend the Summer of 1951 as a cooperator of the Arctic Aeromedical Laboratory at Ladd Air Force Base, Alaska. The following data were gathered incident to the collection of small mammals and their ectoparasites for the laboratory. Observations from June 26th through July 2nd, 1951, were made on Barter Island, which is a small, low-lying bit of land shaped like an isosceles triangle approximately four miles on a side and which lies just off the Alaska mainland in the Beaufort Sea (Lat. 70° 7′ N., Long. 143° 40′ W.). The northern shore corresponds to the base of the triangle with the extreme southern tip as its apex. It is separated from the mainland at the closest point by a slough approximately three-quarters of a mile wide. A low bluff on the north side varies from two to three feet on the east to twenty or thirty feet near its center and western end. A narrow gravel beach from ten to fifty feet wide completely surrounds the island. The peat-tundra soil overlays a sand-gravel substratum, the peat layer varying from only a few inches to at least fifteen feet in thickness.

The ground thawed to an average of only eight inches during our stay, and the early vegetation was just beginning to bloom. Much of the island is thickly covered with low growing tundra plants of relatively uniform composition. Elevation differences of a few feet give the impression of a rolling prairie when viewed from the air. Small depressions holding snow water were numerous, particularly near the northern shore, at which place we captured most of our specimens. Several of the snow water lakes on the island are permanent.

Only four species of small mammals could be definitely established as resident throughout the year. They are: the arctic fox *Alopex lagopus innuitus* (Merriam), the tundra mouse *Microtus oeconomus macfarlani* Merriam, the brown lemming *Lemmus trimucronatus alascensis* Merriam, and the collared lemming *Dicrostonyx groenlandicus rubricatus* (Richardson). The esquimos reported that weasels and shrews occurred on the mainland "toward the mountains" but that none to their knowledge lived on the island. If any of either of these forms were present, they were quite scarce, for we saw no signs of them.

A *Microtus* colony was located on the northeast corner of the island. It appeared to be composed of a number of individuals, but we were successful in securing only thirteen specimens, all adults. The *Microtus* appeared to be re-

stricted to the low-lying peninsula at the site of an abandoned esquimo village (Plate 19, fig. 1) where their runs and tunnels through the vegetation were quite similar to, but smaller than, those of *Lemmus*. The area had been modified considerably from its original condition by the establishment of an airstrip. Prior to the building of the base, the area appeared to have been a low, poorly drained swale with some tundra-covered ridges a foot or so higher than the general marsh level. While some of this habitat still remains, parts of it are better drained and over-all drier conditions now prevail.

The nests discovered consisted of loose, bulky masses of vegetation, the single chamber being lined with fine, often shredded, material. The space available for the nests determined the size and shape of those examined, for the mice had filled every cranny of the chamber completely. *Poa pratensis* var. *alpignea* E. Fries, *Poa arctica* R. Br., and *Stellaria laeta* Richards. were the primary species utilized for nest construction. The seeds of these plants were probably used for food during the winter and spring, for there were few fruits left attached and in several nests an accumulation of "chaff" was distributed throughout the nesting material. While the voles showed a preference for nesting in the spaces between the tundra blocks of the collapsed igloo walls, one nest was found under a weathered skull of a bow head whale (*Balaena mysticetus* Linnaeus), another was discovered beneath a driftwood stump, and several were seen on the ground in the spaces among fifty gallon oil drums which were stacked near the village site. The mice obviously preferred to build their nests in pre-existing cavities and rarely attempted to enlarge or improve them. They demonstrated a marked selectivity for the drum stacks as an escape refuge, for in a number of instances when the tundra blocks or driftwood under which they were hiding were turned over they would scamper with incredible rapidity across several yards of exposed ground to reach the drum pile even though more accessible holes were available nearby. Once they reached the stack, they were afforded almost complete immunity from harm by man or by other enemies. The drum pile provided them with a safe retreat even from the arctic fox, although they were extremely vulnerable to predation while crossing the gravel strip to reach the piles. The only arctic fox reported during our stay was seen in the drum area.

The mice runways were littered with soft, bright green droppings. The majority of these trails led from the old house row to the wet snow-water slough area where the voles were feeding on the new succulent vegetation.

They did not seem to be feeding on the dry grass seeds which were still available in abundance on the higher areas nearby, although they were harvesting *Poa pratensis* var. *alpignea*. In the stands of this species, there were patches a foot or more in diameter in which every culm had been cut close to the ground with the butts oriented in the same direction. It appeared that the mice would cut a quantity of stalks before carrying them away for subsequent use. No seed caches were discovered and the "chaff" found in the old nests suggest that they may construct these structures using the entire stalk with the fruit and leaves attached. The ripened seeds would then be available for winter use and the mice could feed without leaving these well-insulated structures. The species *Microtus*

oeconomus seemed to show a definite preference for the wetter areas wherever we found them and their trails, workings, and other activity signs, reminded us of those made by *Microtus pennsylvanicus nigrans* Rhoads in the coastal marshes of North Carolina.

No nests containing young were found. Of six females examined, two had recently ceased lactating, three contained early embryos (1 to 2 mm. long), and the sixth had produced young sometime previously but was neither lactating nor pregnant at the time of collection. Embryo counts in the pregnant females were eight, nine, and eleven. The female with the nine embryos (1 mm. long), moreover, appeared to have just ceased lactating. The five males examined were sexually mature and were apparently sexually active. The testes dimensions averaged 7.8 mm. long by 5.6 mm. wide. No record was made of the breeding condition of the two *Microtus* kept alive.

These data are insufficient, of course, to determine the presence or absence of a breeding peak in the spring despite the fact that one is indicated. At least a part of this colony had bred in May, at which time there was an abundance of snow still present.

The series of tundra mice obtained on Barter Island are tentatively identified as *Microtus oeconomus macfarlani* Merriam, although they do not agree very closely to Merriam's type. They are more similar to specimens taken near the locality from which Osgood described *M. o. endoecus*. Should *endoecus* prove not to be a synonym of *macfarlani* (Setzer, 1952), the Barter Island series may be referable to *M. o. endoecus* Osgood. The age and condition of Merriam's material and the lack of sufficient additional material from the type locality of *macfarlani* make the author reluctant either to describe his series as a race distinct from *macfarlani* or to recognize *endoecus* as a valid subspecies.

There were no lemmings present in the area occupied by the *Microtus*, and there appeared to be no *Microtus* on the portion of the island occupied by the lemmings. When both the lemming and the vole populations are low, the species probably do not come into contact with each other.

Rausch (1950) recorded a cyclic die-off of both genera of lemmings in the vicinity of Point Barrow during the Spring of 1949. The low may have occurred at Barter Island about the same time, although we could obtain no positive confirmation. The lemming population was apparently still very low both on the island and on the mainland in August, 1950, for R. L. Strecker and P. B. Morrison (1952) were able to secure only two *Lemmus* and three *Dicrostonyx* in five days of intensive work. One of the *Dicrostonyx* they captured was a female with a litter of four.

About two-thirds of our specimens were caught by hand, the remainder being taken in Rinker and Sherman live traps. Bacon, peanut butter, and a combination of the two were used as bait; but, since all specimens were taken in runways, it is believed that the bait had little if anything to do with attracting the animals. Only three collared lemmings (*Dicrostonyx*) were secured, as compared with forty-two brown lemmings (*Lemmus*), during the six days of collecting. The ratio of three *Dicrostonyx* to forty-two *Lemmus* may approach that which existed in the

population at the time, since we searched the entire island for lemming and set traps wherever there was any evidence of a concentration.

Dr. F. Bell caught eight *Lemmus* and one *Dicrostonyx* on the island on July 29th. The collared lemming and the majority of the brown lemmings were subadults. One adult female *Lemmus* gave birth to at least five young shortly after her capture. Only one of Dr. Bell's nine lemmings was trapped, the others being caught by hand.

One of the *Dicrostonyx* we secured was live trapped in an active run where two *Lemmus* had been taken previous to and one after its capture. Another was caught by hand in an active *Lemmus* trail, and the third was taken in an old run, which was possibly made by *Dicrostonyx* despite the fact that several *Lemmus* were caught in the same area. On Southampton Island, Sutton (Sutton and Hamilton, 1932) found both *Dicrostonyx* and *Lemmus* using the same burrows in the winter; while in the summer, the former species tended to live principally among the rocks along ridges, and the latter favored the flat country. The relative uniformity of the habitat on Barter Island made the separation of the two forms into different areas virtually impossible. It was, however, obvious that *Lemmus* trails were most abundant in the wet areas and that their meandering paths led from one pond of snow water to another, usually along the lowest contour. In and around these snow water pools, they were feeding heavily on the new growth of *Eriophorum angustifolium* Rath. and *Carex lugens* Holm, the luxuriant growth of which had the appearance of having been cut by a lawn mower. *Lemmus* were also feeding on *Ranunculus sulphureus* Phipps and were browsing heavily in several areas on roots and underground stems, from tunnels a few inches below the surface of the small hummocks, particularly where *Salix reticulata* Linn., *Salix pulchra* Cham., and *Salix phlebophylla* Anders. were growing.

A number of the summer nests of *Lemmus* were found in these small hummocks, two of which contained nine young approximately one and five days old respectively. It is not surprising that the mothers of these litters were not seen, since the female lemming apparently stays in the nest only long enought to suckle the young and then retires to another chamber nearby. This behavior pattern was observed in several caged brown lemmings which gave birth to young in captivity. The summer nests were globular in shape, usually about six inches in diameter, although some were as small as four inches. They were composed almost entirely of the shredded leaves and stems of sedges, though a few *Salix* twigs were included. Several of the nests were so close to the surface that they could be seen by peering down the short entrance tunnel which provided the single entrance characteristic of all nests examined.

Although several old nests presumably belonging to *Dicrostonyx* were examined, no known nest of this species was found. Those examined were located in the large hummocks, or "owl mounds", which were heavily tunnelled; and, in many instances, runways like miniature highway cuts three inches wide and six to twelve inches deep extended through them (Plate 19, fig. 2). All workings of this nature, and the majority of droppings in them, were quite old. A few *Lemmus* were using these areas, and it was thought at the time that the diggings

represented their winter trails; after reading Sutton and Hamilton's report, however, we believe that these excavations were made by *Dicrostonyx* when their population was at a higher level than at present. All of our observations on known *Lemmus* diggings are in agreement with Sutton's, for their tunnelling was of a more superficial nature than those attributed to *Dicrostonyx*.

Three types of chambers led from *Lemmus* runs or tunnels. The first appeared to be a feeding shaft three to six inches long and often ending in a small globular room only a little larger than the tunnel itself, the room frequently containing piles of fresh droppings. The second type seemed to serve as an escape or resting chamber, for several of them could be found along well-used runs. Whether they were dug while exploring for food or constructed for resting and protection was not determined. In either event, they provided a safe resting site from winged predators. The third type was the nest chamber.

In the Spring of 1949, Rausch examined 139 lemmings, mostly *Lemmus*, prior to and during the time of decline and found no evidence of reproductive activity. Of the 139, only 59 were females, and there were no very young animals in the population.

Of the forty-two adult *Lemmus* we collected, twenty-four were females, seventeen males, and the sex of one was not recorded. All but two of the males were sexually mature. Fourteen of the twenty-four females were pregnant with a total of ninety-eight embryos, embryo counts varying from four to nine, an average of 7.3 each. One female was actively lactating, and two had just ceased. None of these three was pregnant. Two females gave birth to at least six young shortly after their capture. Only three females were neither pregnant nor lactating, and one of these had produced young sometime earlier. Reproductive data on two were lacking. The only female *Dicrostonyx* taken contained nine small embryos. One male was a sub-adult, the other, a sexually mature male.

Sutton and Hamilton reported reproductive activity in *Lemmus* throughout the year on Southampton Island. Preble's (1902) comment on his observations near Churchill from August 4th to 8th, 1900, that the breeding season seemed to be nearly over would indicate that he believed them to have a definite breeding period. Our data seemed to imply at least that breeding activity peaks exist, for slightly over 86 per cent of all females examined were either pregnant or suckling young, no recently weaned *Lemmus* were seen or taken, and only two of the forty-two captured were sub-adults, no other sub-adults being observed.

From January 16th to March 20th, 1930, at which time the lemming population was high, Sutton and Hamilton reported an average of 3.7 embryos (3–5) in eleven pregnancies. He reported a die-off in June of the same year. Litters found in the summer nests were reported by a Mr. Jack Ford to Sutton as nearly always having six or seven young, but these counts were not made during the year Sutton made his study and may have been made during a period of buildup. Preble reported from near Churchill that four to six embryos in an unstated number of pregnancies were found in August, 1900, another year when lemmings were at a peak. Soper (1944) reported one female *Lemmus* with seven embryos taken on Baffin Island on June 30, 1929, during a buildup.

Rausch's observation of no breeding activities prior to and during the cyclic

decline at Point Barrow is, in our opinion, a population characteristic during this period. A high percentage of breeding adults and characteristically large litters may be typical of a cyclic buildup in *Lemmus* in its early stages.

A number of regurgitated snowy owl, *Nyctea scandiaca* (Linnaeus), and jaeger, *Stercorarius* sp., pellets found on the tundra hummocks or "owl mounds" (Plate 19, fig. 2) were examined. *Lemmus* remains made up the entire contents of those identified on the basis of size as being from the snowy owl, while those known to be from the jaegers often contained bird material. Approximately two dozen pellets were examined with the hope of finding evidence of small mammals other than those whose presence was already known, but only the remains of *Lemmus* were found in any of the pellets containing mammalian material.

The owl population was estimated roughly at three birds per square mile, while the jaeger concentration was slightly less. When undisturbed, the owls spent almost all their time on the tundra hummocks or on telephone poles and left their perches only to make a short low flight presumably in search of food. On only one occasion was an owl seen to make a pass at something on the ground. Although we could not be positive, it was believed that its intended victim was a lemming, since no bird was seen to flush and since the owl lit only a few feet beyond its apparent target. The owl then remained completely immobile for several minutes and took wing only after we began to approach. On any sustained flights over the island, the owls were frequently attacked by one and sometimes by a succession of Lapland longspurs (*Calcarius lapponicus*), each one in turn harrassing the clumsy predator as it passed over its territory. Once a male snow bunting (*Plectrophenax nivalis*) was seen to make a vigorous attack on a passing owl. Both the longspurs and the buntings were nesting at the time of our visit. Although there were numbers of shore and passerine birds present on the tundra, the snowy owls seemed to be devoting almost their entire hunting efforts to securing lemmings, for they made little attempt to chase or even to deviate from their course when these small birds flushed in front of them. This situation was attributed not only to the greater vulnerability of the lemmings at this time owing to the absence of snow and to the short vegetation, but also to the fact that some of the carcasses of lemmings which had died during the preceding winter were still available as carrion. The jaegers seemed to be making more use of this fortuitous supply than were the owls; however, some of the *Lemmus* material in the owl pellets also appeared to have been taken as carrion.

The parasitic jaeger, *Stercorarius parasiticus* (Linne), was the most common of the three species of jaegers observed. One pomarine jaeger, *Stercorarius pomarinus* (Temminck), was found dead and one or perhaps two different individuals observed patrolling the tundra were believed to have been this species. Two parasitic jaegers found dead had been partially devoured. One of the birds was discovered when the spot at which a parasitic jaeger was observed feeding was investigated. From the condition of the carcass, the bird appeared to have been the victim of a snowy owl rather than of the jaeger. Several long-tailed jaegers (*Stercorarius longicaudus* Vieillot) were seen actively searching the tundra

for food, one of which was flushed from a hummock where it had perched for several minutes. One freshly regurgitated pellet was found at the perch and several older ones about the same size were also picked up. Whereas *Lemmus* remains were found in nearly every one, bird material made up the bulk of the contents.

The jaegers were on the wing almost constantly and were observed to make a number of unsuccessful attacks on the small birds on the ground, particularly the sandpipers (*Pisobia* sp.). The remains of one longspur was dropped by a jaeger and one sandpiper carcass was found which had been almost completely eaten, presumably by a jaeger. No known attacks on lemmings were observed, but both the parasitic and the long-tailed jaegers were seen making passes at ground targets. From their actions and the fact that no birds flushed from the spot, it was believed that these attacks were directed at lemmings. The longspurs did not hesitate to defend their territories as vigorously against the jaegers as against the snowy owls.

Despite the fact that our period of observation at Barter Island was brief, it was clear that this area would be an ideal one in which to study population dynamics of these small mammals. The few species and the relative uniformity of vegetation and habitat provide a natural experimental area where the number of variables which usually complicate such studies are reduced to a minimum.

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PLATE 19

FIG. 1. Ruins of a tundra block-driftwood igloo, the crevices in the walls were being utilized by a colony of *Microtus oeconomus macfarlani* Merriam.

FIG. 2. A tundra hummock or "owl mound" heavily tunnelled by lemmings. Jaegers and snowy owls often perched on these vantage points. A number of their regurgitated pellets could usually be found on every mound.

THE EFFECT OF ADRENALECTOMY ON NATURAL AND ACQUIRED RESISTANCE OF MICE TO *TRICHINELLA SPIRALIS**

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Trichinella spiralis, a parasitic nematode, is the cause of trichinosis, a disease of public health importance in this country and elsewhere. Much has been learned about resistance to this parasite, but many important questions remain unanswered. Most of the available information on resistance has been derived from studies in experimental animals. Of these, white mice have been used a great deal in recent years, as they are well adapted for use in large experiments in which quantitative measurements are desired. The ultimate goal of most resistance studies is to learn the mechanisms involved. In the case of *T. spiralis*, such knowledge might be of great value in the development of better methods of early diagnosis, treatment and prevention of trichinosis.

One of the most commonly used approaches in attempts to gain an insight into the mechanisms of resistance is to modify the host's resistance by some factor and to study the effect on the parasite. In mice infected with *T. spiralis*, various factors such as alcohol (Larsh and Kent, 1949), a vitamin A deficient diet (Larsh and Gilchrist, 1950), DDD (Baughn, 1950), and concurrent infections (Larsh and Campbell, 1952; Cox, 1952a and b) have been studied. However, no reports have been found dealing with the use of adrenalectomy as a factor to modify resistance to this parasite.

Considering the wide range of physiologic effects produced by adrenalectomy, many of which influence resistance directly or indirectly, it seemed desirable to study the effect of this factor in modifying the resistance of mice to *T. spiralis*. It was decided to study the effect of adrenalectomy both on natural and acquired resistance with the hope that the data obtained might be helpful in designing future experiments to investigate the mechanisms of resistance. The results of the present study have been presented previously in abstract form (Baughn, 1952a, 1952b).

Materials and Methods

White female mice, five weeks of age, were used in this experimental study. The mice were raised in this laboratory and were of the same stock used in

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experiments reported previously by members of this Department (Larsh and Kent, 1949; and others).

Procedure for adrenalectomy.—Animals were adrenalectomized in a two-stage operation. The mice were anesthetized by the intraperitoneal injection of 0.04 c.c.–0.06 c.c. of a 50 per cent solution of Veterinary Nembutal Sodium (Abbott), diluted with 0.85 per cent saline. From three to five minutes were allowed for the nembutal to take full effect, after which the hair over the incision site was clipped short with electric clippers. In all cases, the right adrenal was removed in the first stage of the operation. Using a dorso-lumbar approach and following aseptic techniques, an incision about 1 cm. in length was made immediately posterior to the last rib and at an angle between the last rib and the vertebral column. Following incision, the blood supply to the gland was constricted with forceps; this procedure is believed to cause complete hemostasis. The loose tissue medial to the adrenal gland, any existing fat surrounding the gland, and the surrounding kidney capsule were torn from the kidney. In this manner, not only was the intact adrenal gland removed, but from one-third to one-half of the kidney was decapsulated. The muscle layers were closed by a single nylon suture, and the skin incision was closed at intervals by a continuous suture.

Two days following the removal of the adrenal gland on the right side, the left adrenal gland was removed in a similar manner. The removal of the gland from the left side was best accomplished by making a slightly more posterior and central approach.

Maintenance of adrenalectomized animals.—Giving 0.9 per cent NaCl solution in place of drinking water has been an accepted procedure for many years to aid the survival of adrenalectomized animals. Therefore, one week prior to adrenalectomy, all mice were given this salt solution, which was provided until completion of the second stage of the operation. At this time, it was necessary to provide additional material to maintain the mice. Desoxycorticosterone acetate (DCA), a synthetic adrenal hormone, was used, since it is known to increase the life span without interfering with the resistance mechanisms of the adrenalectomized animals (Ettelson, 1940). This hormone possesses the ability to aid in maintaining the fluid and electrolyte balance, usually upset following adrenalectomy, but it does not correct the disrupted sugar and nitrogen balance. These facts have a bearing on the discussion of the experimental results to be presented in a later section. The DCA used was derived from Percorten Linguets (Ciba).*

To determine the best method of administering DCA and to find the desired quantity for use in the various experiments, it was necessary to make certain preliminary comparisons (Diag. 1). Four mice were used in each of eight groups tested. As shown in Diag. 1, both 10 and 20 per cent DCA in NaCl solution proved very effective in maintaining the mice following adrenalectomy. However, it was decided to use the 10 per cent concentration to minimize the chances of interference with the resistance mechanisms.

Stock strain of T. spiralis, *methods of isolating and administering larvae for*

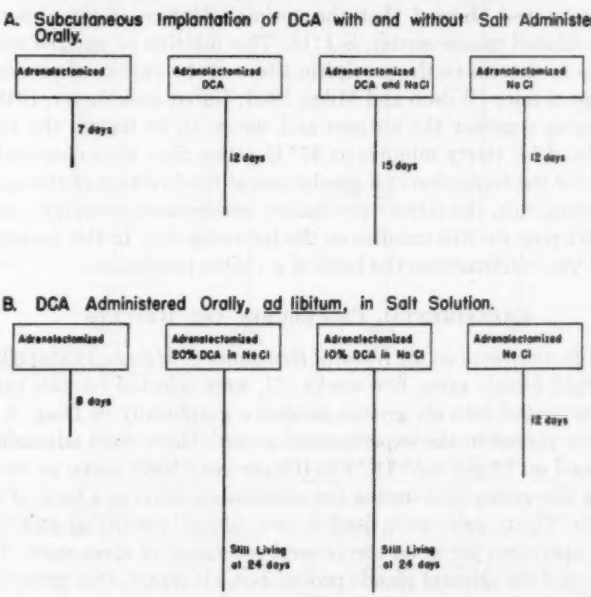
* Supplied through the courtesy of Mr. W. A. Diehl, representative for Ciba Pharmaceutical Production, Inc.

infection, and necropsy procedures.—The stock strain of *T. spiralis* was one that has been maintained in mice for several years. The various experimental procedures were those reported earlier from this laboratory (Larsh and Kent, 1949), except that the larvae for infection were kept at body temperature from the time of isolation until infection.

Measurement of resistance.—For natural resistance, the measurement was the number of adult *T. spiralis* recovered from the small intestine of the mice seven days after infection with 100 larvae. Comparisons were made between the numbers recovered from the various experimental and control groups. In

DIAGRAM I

Design and Results of Preliminary Experiments to Determine Effect of DCA (Percorten Linguets (Ciba)) on Average Longevity of Adrenalectomized Mice.



the case of acquired resistance, the experiment was designed to test the effect of adrenalectomy on the maintenance of the resistance. Therefore, certain of the mice were first immunized after which some were adrenalectomized. Other mice served as non-immunized controls to show the degree of natural resistance of mice of this age. Four days following the adrenalectomies, all of the mice were given a challenging (test) infection with *T. spiralis*. The measurement of the maintenance of acquired resistance was the number of adult *T. spiralis* recovered from the small intestine of mice of the various experimental and control groups seven days after the challenging infection.

The precipitin test.—This test was used in the experiment on acquired resistance to show the relationship of antibody titer to worm burden. Previous work in mice has proved the value of this test (Hendricks, 1950, 1952).

The antigen used in this test was Allergenic Extract Trichinella, a 1:10,000 dilution of an antigen prepared from *T. spiralis* larvae.* To collect the serum for this test, the animals were bled just prior to necropsy. Each animal was anesthetized with nembutal, and the axillary artery of the left front leg was severed and allowed to bleed. The blood was collected by the use of a 1.5 c.c. syringe and placed in a sterile tube. The blood from each group of animals, experimental and control, was pooled in separate tubes and allowed to clot at room temperature. The clot was then removed and the tube refrigerated overnight at 4° C. The following day each tube was centrifuged at 1500 r.p.m. for fifteen minutes and the serum collected. The serum was then stored in a deep-freeze unit until needed.

A preliminary test showed that the optimal dilution of the above antigen, used with undiluted mouse serum, is 1:16. This dilution of antigen was used in the final tests to determine the precipitin titers of the various experimental and control groups of mice (Wilson and Miles, 1946; Kabat and Mayer, 1948).

After bringing together the antigen and serum to be tested, the tubes were allowed to stand for thirty minutes at 37° C.; then they were observed at room temperature for the formation of a precipitate at the junction of the antigen and serum. Following this, the tubes were shaken, refrigerated overnight, and centrifuged at 1500 r.p.m. for five minutes on the following day. In this manner, a positive reaction was confirmed on the basis of a visible precipitate.

EXPERIMENTAL PROCEDURES AND RESULTS

Effect of Adrenalectomy on the Natural Resistance of Mice to Trichinella spiralis. —Seventy-eight female mice, five weeks old, were selected for this experiment. These were arranged into six groups as shown graphically in Diag. 2. Twenty-two mice were placed in the experimental group. These were adrenalectomized and maintained on 10 per cent DCA in 0.9 per cent NaCl given as the drinking fluid. Five of this group died during the experiment, leaving a total of seventeen experimentals. Thirty mice were used in two "sham" control groups to test the effect of the operation *per se* on the natural resistance of these mice. They were operated on, and the adrenal glands probed but left intact. One group of twenty shams received DCA (given as above) after the operation, the other group of ten shams did not receive this hormone. Twenty mice served as regular controls not subjected to the operation as a control on the effect of DCA *per se* on natural resistance. One group of ten regular controls received this hormone, the other group of similar size did not. The last group of six mice was included as a check on the effectiveness of the adrenalectomies. These adrenalectomy survival controls received no DCA or NaCl following adrenalectomy.

* Supplied through the courtesy of Mr. D. F. McGowan, representative for Eli Lilly and Co.

Four days after the adrenalectomies had been performed, some of the mice of each of the above groups, except the adrenalectomy survival controls, were infected with 100 *T. spiralis* larvae. Eleven days after the adrenalectomies had been performed, the remaining mice in all groups, except the adrenalectomy survival controls, were infected with 100 *T. spiralis* larvae. In both cases, seven days after infection the mice were sacrificed and counts were made of the num-

DIAGRAM 2

The Effect of Adrenalectomy on Natural Resistance of Mice to Infection with Trichinella spiralis.

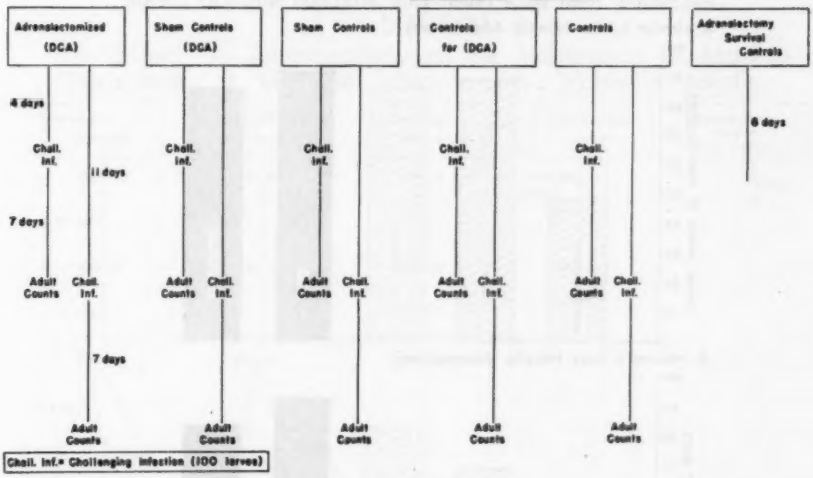


TABLE 1

The average number of adult T. spiralis *recovered from adrenalectomized and control mice seven days after a single infection with 100 larvae*

TIME FROM ADRENALECTOMY TO INFECTION (DAYS)	EXPERIMENTALS (ADRENALECTOMIZED, THEN GIVEN DCA)		SHAM CONTROLS (SHAM OPERATION)				REGULAR CONTROLS (NO OPERATION)			
			Given DCA		No DCA		Given DCA		No DCA	
	No. mice	Ave. no. worms	No. mice	Ave. no. worms	No. mice	Ave. no. worms	No. mice	Ave. no. worms	No. mice	Ave. no. worms
4	7	48.3	8	88.3	4	91.2	4	86.0	4	85.2
11	10	35.8	12	68.3	6	93.1	6	83.3	6	84.3

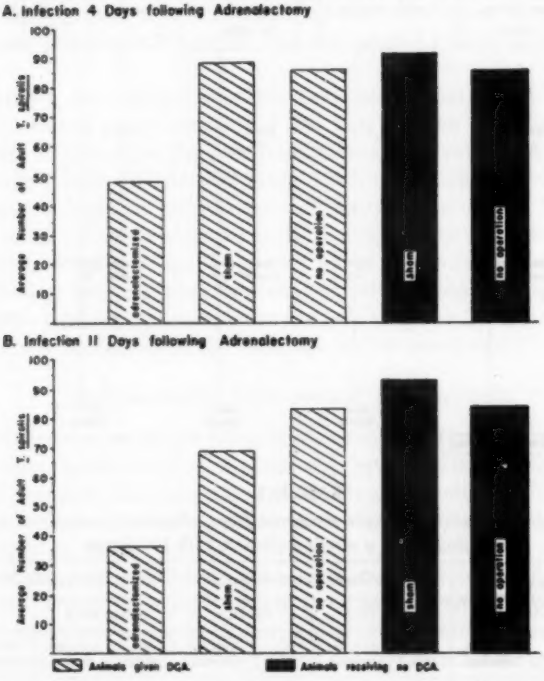
ber of adult *T. spiralis* recovered from the small intestine. The average numbers of worms recovered from mice of the various groups are shown in Table 1, and these findings are shown graphically in Diag. 3.

The effectiveness of the adrenalectomies in both the four-day and eleven-day tests was verified by the early death of the adrenalectomy survival controls (Diag. 2). In the four-day test, the average number of adult *T. spiralis* harbored

by the adrenalectomized mice (48.3) is much lower than that harbored by the shams given DCA (88.3), the shams not given DCA (91.2), the regular controls given DCA (86.0), and the regular controls not given DCA (85.2). An analysis of variance test was performed to indicate the degree of statistical significance of these differences; in this way, the effect of the various factors involved in producing such differences could be determined. (All data are on file, Department of Parasitology, University of North Carolina.)

Diagram 3

The Average Number of Adult T. spiralis Recovered from Mice Given One Infection with 100 Larvae



Analysis showed that the treatments involved in this experiment (adrenalectomy, sham operation and no operation) had a significant effect upon the number of adult *T. spiralis* recovered. Of these treatments, only adrenalectomy was significant in reducing the worm burden at necropsy. This treatment alone was shown to have reduced the recovery by an average of 28.9 worms.

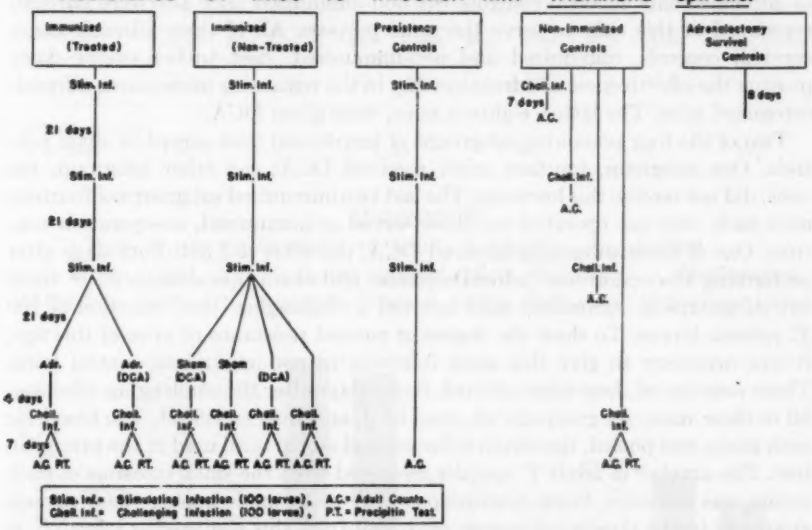
In the eleven-day test, results somewhat similar to those of the four-day test were observed. The average number of adult *T. spiralis* recovered from the adrenalectomized mice (35.8) is considerably less than that from the shams given DCA (68.3), the shams not given DCA (93.1), the regular controls given DCA

(83.3), and the regular controls not given DCA (84.3). It should be noted also that the average number recovered from the shams given DCA (68.3) is much lower than that from the other control groups.

An analysis of variance test showed that the treatments involved (adrenalectomy, sham operation and no operation) had a significant effect upon the number of worms recovered. Again adrenalectomy had a significant effect, having accounted for an average reduction of 36.0 worms. In this case, however, there were two other significant effects, which were not observed in the four-day test. DCA had a significant effect in reducing the number of worms in the non-adrenalectomized mice, and there was a significant interaction effect between the

DIAGRAM 4

The Effect of Adrenalectomy on the Maintenance of Acquired Resistance of Mice to Infection with Trichinella spiralis.



sham operation and the administration of DCA. The latter effect is readily observed by glancing at the worm recoveries from the various non-adrenalectomized mice (Table 1). The shams given DCA showed a much smaller worm recovery (average, 68.3) than the shams not given DCA (average, 93.1). Whether or not DCA was given to the regular controls had little effect on the worm recovery (averages, 83.3 and 84.3). Thus, there was an interaction effect between sham treatment and DCA, which was not shown in the four-day test.

Effect of Adrenalectomy on the Maintenance of Acquired Resistance of Mice to Trichinella spiralis.—One-hundred and eighteen female mice, five weeks old, were selected for this experiment. These were arranged into five main groups as shown in Diag. 4. The mice of three of these groups, eighty-four mice, were im-

munized against *T. spiralis* by giving them three stimulating (immunizing) infections of 100 larvae each at three-week intervals. To check on the viability of the larvae used in these infections, six additional mice, listed under non-immunized controls in Diag. 4, were included at each infection, *i.e.* eighteen mice were used for this purpose. In all cases, the number of adult *T. spiralis* recovered from these "viability controls" seven days after infection was within the range usually observed for this infecting dose. It will therefore be unnecessary to refer again to these controls.

Three weeks after the last (third) stimulating infection, the mice of two of the main immunized groups, seventy-three mice, were distributed into six subgroups (the other main group of immunized mice, persistency controls, will be mentioned below). Two of the six subgroups, twenty-one mice, were adrenalectomized. One of these subgroups, three mice, was not given NaCl or DCA to serve as adrenalectomy survival controls. Six non-immunized mice also were adrenalectomized at this time to serve the same purpose. All of these adrenalectomy survival controls, immunized and non-immunized, died within twelve days, proving the effectiveness of adrenalectomy in the remaining immunized, adrenalectomized mice. The latter, eighteen mice, were given DCA.

Two of the four remaining subgroups of immunized mice served as sham controls. One subgroup, fourteen mice, received DCA; the other subgroup, ten mice, did not receive this hormone. The last two immunized subgroups of fourteen mice each were not operated on; these served as immunized, no-operation controls. One of these subgroups received DCA, the other did not. Four days after performing the operations (adrenalectomies and sham operations), all of these five subgroups of immunized mice received a challenging (test) infection of 100 *T. spiralis* larvae. To show the degree of natural resistance of mice of this age, it was necessary to give this same infection to non-immunized control mice. There were ten of these mice infected. Seven days after the challenging infection, all of these mice, six groups in all, were bled, and then sacrificed. The blood for each group was pooled, the serum collected and stored until used in the precipitin test. The number of adult *T. spiralis* recovered from the small intestine of each mouse was recorded. Since comparisons were to be made among these various groups as to the number of worms recovered from this challenging infection, it was important to know how many worms, if any, had persisted from the last stimulating infection given four and one-half weeks before necropsy. This determination was made by counting the worms from the persistency controls, which received only the three stimulating infections and were sacrificed at the same time as the other mice. An average of only 2.2 worms was recovered from the eleven persistency controls. For this reason, and also because all of the immunized mice received the same stimulating infections, the factor of persistency will be disregarded in the discussion which follows. The results of this experiment, average adult counts and average precipitin titer, are shown in Table 2, and the results of the counts are represented graphically in Diag. 5. The adult counts shown in Table 2 and Diag. 5 are adjusted counts obtained from the actual counts by using the difference in counts for the non-immunized controls in the

present experiment and the counts for the same controls in the four-day test on natural resistance (Table 1). This adjustment was necessary so that a direct comparison could be made between the two experiments.

It will be noted from Table 2 that the adrenalectomized mice harbored a much smaller average number of worms (22.1) than the shams given DCA (48.6), the

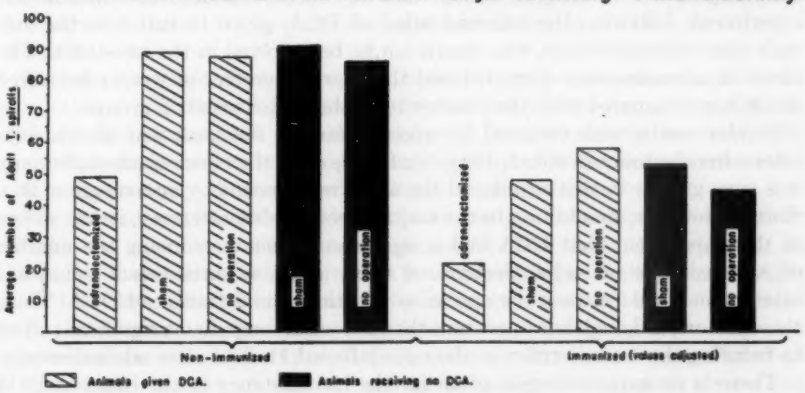
TABLE 2

The average number of adult T. spiralis *and the average precipitin titer of immunized and non-immunized mice following a challenging infection with 100 larvae*

IMMUNIZED MICE										NON-IMMUNIZED MICE	
Experimentals (adrenalectomized)		Sham controls (sham operation)				Regular controls (no operation)				No operation	
DCA		DCA		No DCA		DCA		No DCA		No DCA	
No. mice	Ave. no. worms	No. mice	Ave. no. worms	No. mice	Ave. no. worms	No. mice	Ave. no. worms	No. mice	Ave. no. worms	No. mice	Ave. no. worms
18	22.1	14	48.6	10	53.8	14	58.2	14	45.7	10	85.2
Average precipitin titer.....	1:32	1:32		1:32		1:32		1:32		1:2	

Diagram 5

The Average Number of Adult T. spiralis Recovered from Immunized and Non-Immunized Mice following a Challenging Infection with 100 Larvae Given 4 Days following Adrenalectomy.



shams not given DCA (53.8), the no-operation controls given DCA (58.2), the no-operation controls not given DCA (45.7), and the non-immunized, no-operation controls (85.2). These counts were subjected to an analysis of variance to determine the statistical significance of these differences.

This analysis showed that immunization had a significant effect in reducing

the worm burden at necropsy. In fact, this effect caused an average reduction of 34.1 worms in the immunized mice, compared with the number in the non-immunized mice. The analysis showed further that the treatments involved in this experiment (adrenalectomy, sham operation and no operation) had a significant effect in reducing further the number of adult worms recovered from the immunized mice. Of these treatments, only adrenalectomy had a significant effect in this reduction. It alone reduced the number of adults recovered by an average of 34.2 worms. Since there was no significant interaction effect between immunization and adrenalectomy, *i.e.* one did not significantly affect the value of the other, the effects of the two would be equal to their sum, or a reduction of about 60–70 worms less than the number for the non-immunized, no-operation controls.

The results of the precipitin test, not subjected to statistical analysis, showed no difference in average titer among the immunized mice (1:32). The non-immunized mice, within seven days after the single (challenging) infection, elaborated antibodies which were detected by this test (average titer: 1:2). Serum from stock mice not infected with this parasite gave a negative precipitin reaction.

DISCUSSION

The results of the present study show that young, female, white mice infected with 100 *T. spiralis* larvae four days after adrenalectomy harbored significantly fewer adult worms, and hence had a greater natural resistance, than sham and no-operation controls (Table 1, Diag. 3). The effect of the operation *per se* was shown by statistical analysis to have had no effect on natural resistance in this experiment. Likewise, the administration of DCA, given to maintain the animals after adrenalectomy, was shown not to be involved in the resistance. The effect of adrenalectomy alone reduced the average number of worms recovered by 28.9, as compared with the number from the various control groups.

Similar results were obtained for mice infected in the same way eleven days after adrenalectomy (Table 1, Diag. 3). In this case, the effect of adrenalectomy was even greater in that it reduced the adult worm count by an average of 36.0 worms. However, in addition to the major effect of adrenalectomy, it was shown in this experiment that DCA had a significant effect in reducing the number of worms in the non-adrenalectomized mice, and also there was a significant interaction effect between the operation and the administration of DCA. Thus, these factors, although less important than adrenalectomy, had a significant effect in reducing the worm burden in the mice infected 11 days after adrenalectomy.

There is no apparent explanation for the inconsistency of the effect of DCA and the interaction effect of the operation and DCA, which were absent in the mice infected four days after adrenalectomy and present in those infected eleven days after adrenalectomy. It is possible, however, that DCA had a stimulating effect on various non-specific factors associated with tissue repair which required longer than four days to produce demonstrable effects on resistance. In the regular controls, upon which no operation was performed, the animals given DCA showed adult counts very similar to those not given DCA. It would appear,

therefore, that the operation was the important factor involved in this difference. This apparent lack of interference of DCA in the resistance mechanisms agrees with the finding in other animals (Ettelson, 1940).

Increased natural resistance, as a result of adrenalectomy, is not in accord with the results of certain previous studies involving other agents and hosts. Such studies have shown that adrenalectomy caused a decreased natural resistance to morphine, histamine, bacterial intoxication (Scott, 1923, 1924, 1928), and diphtheria toxin (Belding and Wyman, 1926). Only one reference was found dealing with the effect of adrenalectomy on natural resistance to an animal parasite. In this case, the resistance to *Trypanosoma lewisi* was lowered by this operation (Marmorston-Gottesman *et al*, 1930). Other workers have found no change in natural resistance to morphine or tetanus toxin following adrenalectomy (Ragoff and Ecker, 1926). No reports were found showing an increased natural resistance to an infectious agent following adrenalectomy, as in the present study.

There is no explanation from the present data to account for the increased natural resistance of the adrenalectomized mice. It is known that many physiologic changes occur following adrenalectomy. Perhaps those of greatest concern here are the decreased ability to utilize vitamins (Perla and Marmorston, 1941), the interference with protein and carbohydrate metabolism (White, 1948; Muntwyler, 1940), and the lowering of the blood sugar (Swingle and Remington, 1944). It does not seem likely that these or other known changes would increase the activity of the defensive mechanisms. It is conceivable, however, that one or more of these changes might produce unfavorable conditions for the establishment and/or maintenance of *T. spiralis*. In this respect, it is interesting that this parasite utilizes large amounts of glycogen during maturation (von Brand *et al*, 1951).

The changes mentioned above, and many others, become more pronounced in relation to time following adrenalectomy. In the present study, the favorable effect of adrenalectomy on natural resistance of the mice likewise increased with time. This suggests a relationship between the physiologic changes associated with adrenalectomy and increased resistance to *T. spiralis*.

The present results also showed that the young mice immunized against *T. spiralis* and then adrenalectomized maintained their acquired resistance, as shown by the number of adult worms recovered from a challenging infection given four days after adrenalectomy. Significantly fewer worms were recovered from the adrenalectomized mice and those of the various immunized groups (shams and no-operation controls) than from the non-immunized mice. The effect of immunization was such that an average reduction of 34.1 adult worms was noted, compared with the number of worms in the non-immunized mice. This shows that all of the immunized mice developed acquired resistance as a result of the three simulating infections given prior to the challenging infection. The development of acquired resistance after this immunization procedure agrees with previous work in mice (Culbertson, 1942; Larsh and Kent, 1949; Hendricks, 1950, 1952).

An observation of considerable interest is shown in the comparison of the

various immunized groups in this experiment. The adrenalectomized mice harbored significantly fewer worms than the mice of the other immunized groups. Analysis showed that there was no significant effect of the operation or of DCA upon the number of worms recovered. This shows, as in the experiment on natural resistance, that DCA did not affect the resistance and thus confirms previous work (Ettelson, 1940). The effect of adrenalectomy alone reduced the adult worm recovery by 34.2. Since there was no significant interaction between immunization and adrenalectomy, it can be stated that the combination of the two measures reduced the worm burden by the sum of the two, or approximately 60–70 worms per mouse. Adrenalectomy, therefore, not only failed to break the resistance acquired from previous stimulating infections, but further increased the resistance. These results differ from those for another parasitic organism, *Trypanosoma lewisi*, in which adrenalectomy failed to affect resistance to reinfection (Marmorston-Gottesman *et al*, 1930).

All of the immunized mice, regardless of treatment, showed the same average precipitin titer (1:32) at the time of necropsy. On the other hand, the non-immunized controls, given only the challenging infection and sacrificed seven days later, showed a titer of only 1:2.

It is clear from the results of the present experiment on acquired resistance that adrenalectomy had no effect on the precipitin titer of the mice. It must be recalled, however, that almost all of the antibodies were produced in response to the stimulating infections prior to adrenalectomy. This operation, then, did not influence the concentration of circulating antibodies. This finding agrees with those of other studies showing that adrenalectomy performed at the height of the antibody curve did not affect the antibody titer (Hektoen and Curtis, 1915; Gates, 1918).

The reduction in the number of adults recovered from the immunized mice, as compared with that from non-immunized controls, was related to a greater concentration of circulating antibodies. This relationship of reduced worm burden and increased antibody titer has been noted previously in mice (Hendricks, 1950, 1952). It should be made clear that this relationship does not in any way explain the mechanism of the acquired resistance. That immunity and the formation of antibodies coincide proves nothing about a causal relationship (Marshall, 1948). In fact, the relationship of reduced worm burden and circulating antibodies varied in degree in the present experiment. Despite similar treatment of the various groups of mice during immunization, which resulted in about the same precipitin titer, the adrenalectomized animals, as compared with the other groups of mice, showed a significantly smaller adult worm recovery from the challenging infection. The increased resistance of the adrenalectomized mice was not, therefore, related to a corresponding increase in circulating antibodies. It is possible that a greater concentration of tissue antibodies existed in the adrenalectomized mice, but no attempt was made here to make this determination.

It is interesting to point out that the reduction of worms resulting from adrenalectomy was about the same in the experiment on acquired resistance (aver-

age, 34.2) as in the experiments on natural resistance (average, 28.9 and 36.0). Thus, the factor(s) involved in reducing the number of worms had about the same effects in the immunized and non-immunized mice. If this factor(s) could be discovered by further research, it might shed light on the physiology of the parasite and give an insight into the true mechanism of this resistance. As an aid in such studies, it would be desirable to know the effect of adrenalectomy on the development of acquired resistance, which could not be included in the present study.

SUMMARY

This study involved the use of white female mice five weeks old at the beginning of the experiments. The experimental mice were adrenalectomized in a two-stage operation and maintained on DCA, a synthetic adrenal hormone. Other groups of mice of the same age were used to control the various factors in the experiments.

In the experiments on natural resistance, some of the mice of the various groups were infected with *T. spiralis* four days after the experimental mice had been adrenalectomized; the remaining mice were infected eleven days after the adrenalectomies had been performed. In both the four-day test and the eleven-day test, the adrenalectomized mice harbored significantly fewer adult worms than mice of the various control groups. In both tests, analysis showed that adrenalectomy was the major effect in accounting for this increased resistance of the experimental mice.

In the experiment on acquired resistance, the experimental mice and various groups of controls were immunized by three stimulating (immunizing) infections. Following immunization, the experimentals were adrenalectomized and maintained on DCA. The various immunized control mice were divided into subgroups to control the factors in the experiment. All of the immunized mice were then given a challenging (test) infection with *T. spiralis*. Non-immunized mice of the same age were given this same infection to show the degree of natural resistance of mice of this age. Seven days later, counts were made of the adult worms recovered from the small intestine, and a precipitin test was performed with sera pooled separately from each group of mice.

All of the immunized mice developed acquired resistance, as shown by significantly lower worm burdens than were observed for the non-immunized mice. Also, the immunized mice had a much greater concentration of circulating antibodies. Analysis showed that adrenalectomy had a significant effect in reducing the worm burden of the experimental mice below the level reached by immunization. Therefore, adrenalectomy not only failed to interfere with the resistance developed prior to adrenalectomy, but it further increased resistance.

The effect of adrenalectomy in increasing resistance was significant in the experiments on both natural and acquired resistance, showing that a common factor(s) was operating. The demonstration of the factor(s) involved might shed light on the mechanism of this resistance.

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THE EFFECT OF CONCURRENT INFECTION WITH THE DOG HOOKWORM, *ANCYLOSTOMA CANINUM*, ON THE NATURAL AND ACQUIRED RESISTANCE OF MICE TO *TRICHINELLA SPIRALIS**

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Much has been learned about resistance to *Trichinella spiralis* from studies in experimental animals, but the mechanisms involved have not been determined. In attempts to gain an insight into these mechanisms, many studies have been made in which the resistance of the host was modified to test the effect on the parasite. In mice, factors such as alcoholic debilitation (Larsh and Kent, 1949), a vitamin A deficient diet (Larsh and Gilchrist, 1950), and adrenalectomy (Baughn, 1952a, 1952b) have been tested. However, little attention has been given to the effect of concurrent infection on resistance to *T. spiralis*.

The few references found that deal with the effect on resistance of concurrent infection in mice show conflicting results. For example, resistance of mice to *Hymenolepis nana* var. *fraterna* was reduced in the presence of a concurrent infection with *Strongyloides* or paratyphoid organisms (Brumpt, 1933; Hunninen, 1936), while such resistance was elevated in the presence of *Nippostrongylus muris* (Larsh and Donaldson, 1944). These differences in results may be found to be due to the action of different defensive mechanisms, or quantitative differences of the same mechanisms. Only one reference was found dealing with a study on concurrent infection in mice in which quantitative measurements were made of the effect on resistance to *T. spiralis* (Larsh and Campbell, 1952). It was decided, therefore, that the present study was desirable, and it was hoped that the results might be helpful in designing future studies on the mechanism of resistance to *T. spiralis*. The dog hookworm, *Ancylostoma caninum*, was chosen as the concurrent agent, as its life cycle in mice makes it particularly suitable for such studies on resistance (Kerr, 1936, 1938). Tests were made of the effect of this agent on both the natural and acquired resistance of mice to *T. spiralis*. These results have been presented previously in abstract form (Cox, 1952a, 1952b).

Materials and Methods

White laboratory mice were chosen as the experimental host for this study, as they are economical and easily handled in large experiments. They have been

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shown to be suitable for resistance studies on *Trichinella spiralis* (Larsh and Kent, 1949; and others), and *Ancylostoma caninum* (Kerr, *l.c.*). For the present study, male mice 8–12 weeks old were selected. In raising and maintaining these mice, every reasonable precaution was taken to prevent infections that might affect their resistance.

The concurrent infectious agent, *A. caninum*, was from a stock that has been maintained in mongrel dogs by this Department. Larvae for infection were obtained from moist charcoal cultures of dog feces containing large numbers of eggs. After 7–10 days' incubation at room temperature, the top one-half inch of each culture was placed in a Baermann apparatus and the filariform (infective) larvae were collected. The larvae were suspended in tap water and the doses for infection were standardized by methods similar to those used in standardizing doses of *T. spiralis* (Larsh and Kent, *l.c.*). The volume of the suspension was adjusted so that 0.05 c.c. contained 400 larvae. The infecting dose of larvae was given orally with a 1 c.c. Tuberculin syringe equipped with a blunted, 18 gauge needle. The needle was inserted into the retropharynx of the mouse and the larvae were forced down the esophagus. Subcutaneous infections were made with a 1 c.c. Tuberculin syringe equipped with a 22 gauge hypodermic needle. The larvae were injected beneath the loose skin at the nape of the neck.

The number of *A. caninum* larvae recovered from the small intestine, liver and lungs of the mice was determined quantitatively in the preliminary experiments. To recovery larvae from the intestine, this tissue was first treated with 0.05% NaOH solution (Larsh and Kent, 1949); then the larvae were concentrated by centrifugation into a 2 c.c. volume and counted directly. This process was shown to remove all larvae, as none was found after complete digestion of the tissue. The liver and lungs of the mice were digested at 37° C. with a 0.7% pepsin–1% HCl mixture (Larsh and Gravatt, 1948). Following complete digestion, the larvae from each organ were concentrated into a 2.0 c.c. volume and counted. In the final experiments, *A. caninum* infections were evaluated qualitatively by gross examinations of the lungs and by microscopic examinations of pressed lung sections of control mice infected two days previously. The former procedure gives an indication of the damage produced by the larvae, while the latter procedure shows roughly the number of larvae involved.

The test agent, *T. spiralis*, was a strain that has been maintained in mice by members of this Department. Isolation and standardization of the larvae for infection were performed by methods described previously (Larsh and Kent, *l.c.*). The mice were infected with a 1 c.c. Tuberculin syringe equipped with a blunted, 18 gauge needle. The needle was inserted into the retropharynx and the larvae were forced down the esophagus. To avoid chilling the larvae during standardization and infection, they were kept in a water jacket adjusted to 37° C. The number of adult worms, or larvae, recovered at a given time post-infection was used as the criterion of resistance. All of the adult worms recovered from the small intestine were counted directly, following overnight treatment of the intestine with the NaOH solution, but dilution methods were used to estimate the number of larvae present (Larsh and Kent, *l.c.*).

The data of the final experiments were analyzed statistically. The data of the four experiments on natural resistance (Tables 3 and 4) were analyzed by Student's "t" test (Fisher, 1946). In these comparisons, a difference significant at the five per cent level was accepted as valid, *i.e.* not due to chance occurrence. The two experiments on acquired resistance (Table 5) were analyzed by the analysis of variance test. (Calculations and tables of these analyses are on file, Department of Parasitology, University of North Carolina.)

EXPERIMENTAL PROCEDURES AND RESULTS

A. *Preliminary Experiments*

Before employing the dog hookworm, *Ancylostoma caninum*, as the concurrent infectious agent in the final experiments, certain information was needed, such as the maximum sublethal dose, etc. It was therefore necessary to perform certain preliminary experiments. The mice of these studies were males eight to ten weeks old.

The first preliminary experiment was conducted to determine the maximum sublethal dose of larvae. Ten groups of 5–6 mice each were used. Five groups received oral infections of various sizes, the other five groups were given the various doses subcutaneously. In the latter case, somewhat larger doses were used, since previous work had shown greater tolerance to subcutaneous infection (Kerr, 1936). The mice were observed twice daily and a record kept of deaths that occurred within a twenty-three day period. The infecting doses of larvae used and the results of this experiment are shown in Table 1.

It will be noted that the only groups of mice in which there were no deaths were those given 800 larvae orally and those given 1000 larvae subcutaneously. These doses were therefore accepted as the maximum sublethal doses for our mice of this age.

Another preliminary experiment was carried out to study the route of migration of the filariform larvae, and the time factors involved following oral and subcutaneous inoculation. Two groups of fifty mice each were used. Maximum sublethal doses were used, so that one group received 800 larvae *per os*, the other received 1000 larvae subcutaneously. At two-day intervals after infection, five mice from each group were sacrificed. In consideration of the usual migration of these larvae, counts were made of the number of larvae in the intestine, liver and lungs (Table 2).

It will be noted that only a small number of larvae was recovered. There are at least two reasons for this. Perhaps the most important, is that the mouse is an abnormal host for this parasite. Also, during the migration from intestine to the lungs and from the lungs back to the digestive tract, the larvae do not remain for long in most organs. The period spent in the lungs is the longest, hence the counts for that organ should be most reliable.

Following oral infection, most of the larvae had left the small intestine within two days, although a few larvae persisted for about one week. About the same results were obtained for the liver, hence this organ is not an efficient barrier to

TABLE 1

The survival rate of mice infected with Ancylostoma caninum

DAYS AFTER INFECTION	NO. DEATHS AFTER ORAL INFECTION WITH VARIOUS NUMBERS OF LARVAE					NO. DEATHS AFTER SUBCUTANEOUS INFECTION WITH VARIOUS NUMBERS OF LARVAE				
	800 (5 mice)	900 (5 mice)	1000 (5 mice)	1100 (6 mice)	1200 (6 mice)	1000 (6 mice)	1200 (6 mice)	2000 (6 mice)	2500 (6 mice)	3000 (6 mice)
1	0	0	0	0	0	0	0	0	0	1
2	0	0	0	0	0	0	0	0	2	4
3	0	0	0	0	0	0	0	0	1	0
4	0	0	0	0	0	0	0	0	1	0
5	0	0	0	0	0	0	0	0	1	0
6	0	0	0	0	1	0	0	0	1	1
7	0	2	0	0	2	0	0	0		
8	0	0	0	3	1	0	0	0		
9	0	0	0	0	0	0	0	1		
10	0	0	0	0	0	0	0	0		
11	0	1	1	1	0	0	0	1		
12	0	0	1	0	0	0	0	0		
13–14	0	0	0	0	0	0	0	0		
15	0	0	0	1	0	0	0	1		
16	0	0	1	0	0	0	0	0		
17	0	0	1	0	0	0	1	1		
18–23	0	0	0	0	0	0	0	0		
No. dead...	0	3	4	5	4	0	1	4	6	6
Per cent dead.....	0.0	60.0	80.0	83.3	66.6	0.0	16.6	66.6	100.0	100.0

TABLE 2

The average number of Ancylostoma caninum *larvae recovered from mice at various intervals after infection (for each day of necropsy, five mice from each infection group were sacrificed)*

NECROPSY PERFORMED AFTER INFECTION (DAYS)	AVERAGE NUMBER LARVAE RECOVERED FROM MICE GIVEN 800 LARVAE ORALLY			AVERAGE NUMBER LARVAE RECOVERED FROM MICE GIVEN 1000 LARVAE SUBCUTANEOUSLY		
	Small intestine	Liver	Lungs	Small intestine	Liver	Lungs
2	5	7	106	0	0	159
4	3	2	27	0	0	27
6	3	3	35	0	1	28
8	2	1	16	0	1	9
10	1	0	1	1	0	2
12	0	1	2	1	0	1
14	1	0	1	0	0	0
16	0	0	1	1	0	1
18	0	0	0	0	0	0
20	0	0	0	0	0	0

the migration in this host. It may be, however, that larvae by-pass the liver and reach the lungs via the lymphatic circulation. Comparatively large numbers were found in the lungs as early as two days post-infection. There was a consider-

able reduction in the number of larvae in this organ by the end of four days, although many persisted for eight days. Following subcutaneous infection, very few larvae were recovered from the intestine and liver, as expected, but the number from the lungs was somewhat similar to that in the mice infected orally. The few larvae found in the small intestine after ten days presumably reached there following migration from the lungs, as in the normal host.

These results indicate that for our mice of this age and sex the maximum sublethal dose of *A. caninum* filariform larvae is an 800 dose given orally or a 1000 dose given subcutaneously (Table 1). Also from these results, and from a knowledge of *T. spiralis* migration, it may be said that there are at least two organs in which both parasites reside for a time, the intestine and the lungs. In the intestine, the maximum damage caused by *A. caninum* larvae given orally occurred within two days post-infection, while negligible damage resulted in this location following subcutaneous infection. As shown in Table 2, few larvae were found in the intestine two days following oral infection, hence the larvae must penetrate the intestinal wall very rapidly. Thus, severe damage results from large numbers of larvae passing through this tissue within a brief period of time. In the lungs, the period of maximum damage following either oral or subcutaneous infection may be set between two and ten days. These periods of greatest damage may be referred to as the "acute" stage of the *A. caninum* infection. Following this stage in the intestine and lungs, the damage was repaired gradually, until at 10 days after infection (intestine) or 20 days (lungs) the organs appeared to be approaching normal appearance. These latter periods may be referred to as the "chronic" stage of the *A. caninum* infection. In the final experiments, presented below, measurements were made of the effect of *A. caninum* larvae, during both the acute and chronic stages, on the natural and acquired resistance of mice to *T. spiralis*.

B. *Final Experiments:* 1. *The effect of an* Ancylostoma caninum *infection on natural resistance of mice to* Trichinella spiralis

In all of the final experiments, the doses of *A. caninum* were set at 800 for oral inoculation and 800 for subcutaneous injection. The latter was reduced from the 1000 level established as the maximum sublethal dose (Table 1), since it was desirable, for two reasons, to have the doses of equal size. This facilitated standardization of the larvae, and, more important, it was necessary to provide the same degree of antigenic stimulation throughout, in case cross immunity might prove to be a factor in the results obtained. The viability of the *A. caninum* larvae used was checked by the inclusion of additional mice, "viability controls". Two such controls were added to each group of mice infected with *A. caninum*. These controls were sacrificed two days post-infection. In all cases, the appearance of the lungs, and the number of larvae observed in pressed lung sections, were typical for the doses of 800 organisms used.

The first two experiments on natural resistance were designed to test the effect of concurrent infection on resistance to the adult phase of *T. spiralis*. The *A. caninum* infections were given two or ten days prior to those of *T. spiralis*, to

determine the effect on resistance to *T. spiralis* in the intestine during the acute and chronic phases, respectively, of the *A. caninum* infection. The last two experiments on natural resistance were designed to test the effect of *A. caninum* on resistance to the larvae of *T. spiralis*. In these cases, the *A. caninum* infections were given ten or twenty days before those of *T. spiralis*, to determine the effect on resistance to *T. spiralis* in the lungs during the acute and chronic phases, respectively, of the *A. caninum* infection.

EXPERIMENT 1.—*The effect on resistance to adult* T. spiralis *of an* A. caninum *infection given two days before the* T. spiralis.—Three groups of mice were used. Eight were placed in one experimental group and infected orally with 800 *A. caninum* larvae, seven were placed in a second experimental group and were infected with 800 *A. caninum* larvae subcutaneously, and fifteen, not infected with *A. caninum*, were used as untreated controls. Two days after the experimentals received the *A. caninum* infections, all of the mice of the three groups were infected with 240 *T. spiralis* larvae. Seven days later, the mice were sacri-

TABLE 3

The average number of adult T. spiralis *recovered from mice given a single infection with* T. spiralis *after receiving one infection with* A. caninum

EXP. NO.	DAYS AFTER *A. caninum* INFECTION	INFECTING DOSE OF *T. spiralis*	EXPERIMENTALS (800 *A. caninum* PER OS)		EXPERIMENTALS (800 *A. caninum* SUBCUTANEOUS)		CONTROLS (NO TREATMENT	
			No. mice	Ave. no. adult *T. spiralis*	No. mice	Ave. no. adult *T. spiralis*	No. mice	Ave. no. adult *T. spiralis*
1	2	240	8	96.5	7	156.8	15	190.8
2	10	280	6	23.5	7	41.0	16	46.1

ficed and counts were made of the number of adult *T. spiralis* recovered from the small intestine (Table 3).

The average number of adult *T. spiralis* recovered from the experimentals infected orally with *A. caninum* (96.5) is significantly lower than that from the experimentals infected with *A. caninum* subcutaneously (156.8) and that from the untreated controls (190.8). The latter two averages do not differ significantly. It is concluded, therefore, that oral inoculation, but not subcutaneous inoculation, of *A. caninum* two days prior to *T. spiralis* increased the natural resistance of the mice to *T. spiralis*.

EXPERIMENT 2.—*The effect on resistance to adult* T. spiralis *of an* A. caninum *infection given ten days before the* T. spiralis.—In this case, six mice were infected orally with 800 *A. caninum*, seven were given a similar infection subcutaneously, and sixteen served as untreated controls. Ten days after the *A. caninum* infection, all of the mice received 280 *T. spiralis* larvae. The mice were sacrificed seven days later and counts were made of the number of adult *T. spiralis* recovered from the small intestine (Table 3).

The average number of adult *T. spiralis* from the mice infected orally with *A. caninum* (23.5) is significantly lower than that from those infected subcu-

taneously with *A. caninum* (41.0) and that from the untreated controls (46.1). The latter two averages do not differ significantly. The conclusion is therefore the same as that above for the two-day test, *viz.* that oral infection with *A. caninum* ten days before *T. spiralis*, but not subcutaneous infection, increased the natural resistance of the mice to *T. spiralis*.

It will be noted that the percentage development of *T. spiralis* differed greatly in Experiments 1 and 2, despite the use of similar infecting doses. The controls of Experiment 1 had an average of 190.8 worms (79.5 per cent of the infecting dose), while those of Experiment 2 had an average of only 46.1 (16.5 per cent). The usual range in percentage of development in our mice is 40.0 to 70.0, hence the above results for Experiment 2 are much below the usual findings. This, however, does not invalidate the results, since all groups received the same dose of larvae. It may be that extreme agitation of the larvae, resulting from difficulty in standardizing the infecting dose, was the cause of their reduced infectivity. This condition was also encountered in Experiment 3 presented below.

EXPERIMENT 3.—*The effect on resistance to* T. spiralis *larvae of an* A. caninum *infection given ten days before* T. spiralis.—Ten experimentals were infected

TABLE 4

The average number of T. spiralis *larvae recovered from mice given a single infection with* T. spiralis *after receiving one infection with* A. caninum

EXP. NO.	DAYS AFTER *A. caninum* INFECTION	INFECTING DOSE OF *T. spiralis*	EXPERIMENTALS (800 *A. caninum* PER OS)		EXPERIMENTALS (800 *A. caninum* SUBCUTANEOUS)		CONTROLS (NO TREATMENT)	
			No. mice	Ave. no. *T. spiralis* larvae	No. mice	Ave. no. *T. spiralis* larvae	No. mice	Ave. no. *T. spiralis* larvae
3	10	240	10	3,211.2	7	1,236.1	11	10,547.4
4	20	280	10	66,477.0	5	45,640.0	11	70,633.0

orally with 800 *A. caninum*, and seven were infected subcutaneously. Eleven mice served as untreated controls. Ten days after the *A. caninum* infections, all of the mice were given 240 *T. spiralis* larvae. Viability controls killed seven days later showed an average adult *T. spiralis* count of only 16 per cent of the infecting dose. It is probable that this low infectivity of the infecting larvae in this experiment was due to factors related to agitation during standardization, as mentioned above. Forty days after infection with *T. spiralis*, the mice were sacrificed, and an estimate was made by dilution counts of the total number of *T. spiralis* larvae harbored by each mouse (Table 4).

The average number of *T. spiralis* larvae recovered from the experimentals infected orally (3,211.2) and the average number from those infected subcutaneously with *A. caninum* (1,236.1) are significantly less than that from the untreated controls (10,547.4). The average from those infected with *A. caninum* subcutaneously (1,236.1) is significantly less than that from the mice infected orally with *A. caninum* (3,211.2). Therefore, both oral and subcutaneous inoculation with *A. caninum* larvae ten days prior to infection with *T. spiralis* caused an increased resistance to *T. spiralis* larvae. Greater resistance resulted from *A. caninum* larvae given subcutaneously than from the same number given orally.

EXPERIMENT 4.—*The effect on resistance to* T. spiralis *larvae of an* A. caninum *infection given twenty days prior to* T. spiralis.—Ten experimentals received 800 *A. caninum* orally and five received the same dose subcutaneously, while eleven mice, untreated controls, received no infection with *A. caninum*. Twenty days later, all of the mice were infected with 280 *T. spiralis* larvae. The viability controls, in this case, harbored the usual number of adult *T. spiralis* seven days post-infection. Forty days following the *T. spiralis* infection, the animals were sacrificed and the total number of larvae per mouse was estimated by dilution counts (Table 4).

The average number of *T. spiralis* larvae harbored by the mice infected orally with *A. caninum* (66,477) is not significantly different from that harbored by the untreated controls (70,633), but the latter average is significantly greater than that for the mice infected subcutaneously with *A. caninum* (45,640). The latter average is not significantly different from that for the mice infected orally with *A. caninum* (66,477). Thus, within this period, the oral infection with *A. caninum* had no demonstrable effect on resistance to the larvae of *T. spiralis*, whereas the dose of the same size given subcutaneously caused an increased resistance.

2. *The effect of an* Ancylostoma caninum *infection on the maintenance of acquired resistance to* Trichinella spiralis

EXPERIMENT 1.—*The effect on acquired resistance to* T. spiralis *of an* A. caninum *infection given two days prior to challenging the resistance to* T. spiralis.—Four main groups of mice were used. The mice of three of these groups, 37 mice, were immunized against *T. spiralis* by giving them three stimulating (immunizing) infections of 200 larvae each at three-week intervals. The mice of the other main group, nine mice, were not immunized. These non-immunized mice were needed to show the degree of natural resistance of mice of this age. At each stimulating infection, three additional mice, viability controls, were infected. In all cases, the number of adults recovered from these mice seven days post-infection was within the usual range for this dose. Three weeks after the final (third) stimulating infection, the 37 immunized mice were distributed into two experimental groups and one control group. One group of experimentals, fourteen mice, was given 800 *A. caninum* larvae by mouth, the other experimentals, eight mice, received this dose subcutaneously. The remaining immunized group, fifteen mice, served as untreated controls. Additional viability controls were infected with the same dose of *A. caninum* larvae, some orally, others subcutaneously. The appearance of the lungs, and the number of larvae observed in pressed lung sections two days post-infection, were typical for the doses used. The nine mice of the fourth main group, the non-immunized mice, were not infected with *A. caninum*; hence they served as non-immunized, untreated controls.

Two days after the *A. caninum* infections had been given, all of the mice were given a challenging (test) infection of 400 *T. spiralis* larvae. Seven days later, the animals were sacrificed and counts made of the number of adult *T. spiralis* recovered from the small intestine. These results are listed in Table 5.

An analysis of variance test showed that the average number of adult *T. spiralis* recovered from the immunized, untreated controls (135.0) is significantly less

than that from the non-immunized, untreated controls (227.0). Thus, immunization had a significant effect in reducing the number of worms at necropsy. This shows that acquired resistance had been developed, and confirms previous work in mice (see below). This statistical instrument showed further that the treatments in this experiment (oral *A. caninum*, subcutaneous *A. caninum*, and no treatment) also had a significant effect in reducing the worm burden at necropsy. The average number of adults from the mice treated orally with *A. caninum* (50.8) is significantly less than that from those treated subcutaneously with *A. caninum* (135.3) and that from the untreated controls (135.0). The latter two values do not differ significantly. Thus, only oral *A. caninum* treatment had a significant effect in reducing the worm burden of the immunized mice below the level of reduction caused by immunization. Since there was no interaction between immunization and the treatments involved, their combined effects brought about the reduction in adult worms noted in the immunized mice treated orally with *A. caninum* (average, 50.8), as compared with the number in the non-

TABLE 5

The average number of adult T. spiralis *recovered from immunized and non-immunized mice given a challenging infection with 400* T. spiralis *larvae after receiving one infection with* A. caninum

EXP. NO.	DAYS AFTER *A. caninum* INFECTION	MICE IMMUNIZED AGAINST *T. spiralis*						MICE NOT IMMUNIZED AGAINST *T. spiralis*	
		Experimentals (800 *A. caninum* per os)		Experimentals (800 *A. caninum* subcutaneous)		Controls (no treatment)		Controls (no treatment)	
		No. mice	Ave. no. *T. spiralis* adults	No. mice	Ave. no. *T. spiralis* adults	No. mice	Ave. no. *T. spiralis* adults	No. mice	Ave. no. *T. spiralis* adults
1	2	14	50.8	8	135.3	15	135.0	9	227.0
2	10	14	106.1	8	152.4	14	172.0	9	260.0

immunized, untreated controls (average, 227.0). The effects of immunization and oral treatment were quite similar, each causing a reduction of about 80 worms.

These results show that infection with *A. caninum*, after acquired resistance had been established against *T. spiralis*, did not interfere with the resistance. In fact, oral treatment with *A. caninum* increased the resistance against *T. spiralis*.

EXPERIMENT 2.—*The effect on acquired resistance to* T. spiralis *of an* A. caninum *infection given ten days before challenging the resistance to* T. spiralis.—The design of this experiment was the same as that of the previous one, except for the time of the *A. caninum* infection. The viability controls for the stimulating infections of *T. spiralis* and the one *A. caninum* infection harbored the usual numbers of parasites for the doses employed. Three weeks after the third stimulating infection had been given to immunize certain of the mice against *T. spiralis*, fourteen of the immunized mice were given 800 *A. caninum* larvae by mouth, eight received this dose subcutaneously, while fourteen immunized mice not infected

with *A. caninum* served as immunized, untreated controls. Nine mice were neither immunized nor given *A. caninum*, thus serving as non-immunized, untreated controls. Ten days after *A. caninum* had been administered, all of the mice received a challenging infection of 400 *T. spiralis* larvae, and seven days later they were sacrificed. Counts were then made of the number of adult *T. spiralis* in the small intestine. These findings are shown in Table 5.

An analysis of variance test revealed that the results were quite similar to those of Experiment 1. The average number of adult *T. spiralis* harbored by the immunized, untreated controls (172.0) is significantly less than that of the non-immunized, untreated controls (260.0), showing that immunization had a significant effect in reducing the worm burden at necropsy. The treatments involved also had a significant effect in reducing the number of worms in the immunized mice. The average number of adults from the mice treated orally with *A. caninum* (106.1) is significantly less than that from those treated subcutaneously (152.4) and that from the untreated controls (172.0). The latter two averages do not differ significantly. Therefore, only oral treatment, among these three treatments in the experiment, had a significant effect in reducing the worm burden below the level of reduction produced by immunization. Since there was no interaction between immunization and the treatments involved, it may be said that the combined effects of immunization and oral treatment reduced the average worm burden of 260.0, noted for the non-immunized, untreated controls, to 106.1, noted for the immunized mice treated orally with *A. caninum*.

Using the same statistical instrument as above, a final analysis was made to determine whether the results of the two-day test differ significantly from those of the ten-day test (Table 5), *i.e.* whether time was a significant factor in the results obtained at the two periods. Before undertaking this analysis, it was first necessary to make certain that such a comparison could be made without violating the assumption of a homogeneous experimental error. Bartlett's test for homogeneity (Fisher, 1946) showed that common variance occurred in both the two-day and ten-day experiments. Also, the mean numbers of adults recovered from the non-immunized, untreated controls in the two experiments do not differ significantly. These two analyses made it permissible to proceed with the analysis to compare the results of the two periods of time.

The average number of adult *T. spiralis* recovered from the immunized mice treated orally with *A. caninum* in the two-day test (50.8) is significantly lower than that from the same group in the ten-day test (106.1). The remaining comparisons of the other groups in the two- and ten-day tests (Table 5) showed no significant difference in the values. Since the effect of oral treatment in reducing the worm burden was greater in the two-day test than in the ten-day test, a diminution of the effect with time is indicated. Therefore, time was an important factor in determining the results of Experiments 1 and 2.

DISCUSSION

From the results of this study it is concluded that, under the conditions of the experiments described, the natural and acquired resistance of the mice to *Trichinella spiralis* was not reduced by concurrent infection with the dog hook-

worm, *Ancylostoma caninum*, as might have been expected, but rather, under certain conditions, was increased significantly, as indicated by the number of *T. spiralis* adults or larvae recovered from a test infection in concurrently infected mice, compared with the number recovered from control animals.

Increases in both natural and acquired resistance to adult *T. spiralis* were demonstrated against test infections initiated two and ten days after oral infection with *A. caninum*, but such increases were not shown against test infections given two and ten days after subcutaneous infection with *A. caninum* (Tables 3 and 5). It is important to recall that after oral infection with the dose of 800 *A. caninum*, but not after subcutaneous infection with this dose of the concurrent agent, the small intestine is damaged severely. The situation is different in measuring the resistance to the larvae of *T. spiralis*. In this case, both oral and subcutaneous infections with *A. caninum* involve the lungs, and the larvae of *T. spiralis* must pass this organ in migrating to the musculature. Increases in natural resistance to the migrating larvae of *T. spiralis* were demonstrated against a test infection initiated ten days after oral and subcutaneous infections with *A. caninum* (Table 4). However, after a *T. spiralis* infection given twenty days after *A. caninum*, there was an increased resistance to the migrating larvae of *T. spiralis* only in mice infected subcutaneously with *A. caninum* (Table 4). There is no explanation for this difference in effect, since, as stated above, the lungs are involved following both routes of inoculation of hookworm larvae. It may be that the lung damage resulting from subcutaneous inoculation of larvae is somewhat greater and persists longer than that following oral infection. There was some indication of this in one of the preliminary experiments (Table 2).

The above results allow the conclusion that the demonstrated effect of concurrent infection in increasing resistance to *T. spiralis* was not the product of cross immunity between *A. caninum* and *T. spiralis*. The effect was shown to be operative in the small intestines of mice two days after *A. caninum* infection, which is too brief a period to allow the development of acquired resistance to *A. caninum*. Further, the effect was shown to be subsiding within ten days, rather than increasing as would be expected if acquired resistance to *A. caninum* were involved. Finally, the effect was evident only when an organ was invaded by both parasites. If cross immunity were operative, one would expect acquired resistance to *A. caninum* to be active regardless of the route of inoculation.

A study of the mechanisms involved in the effects produced by the hookworm larvae was not a part of the present experimental design, but the local inflammatory response to *A. caninum* is used as a basis for a hypothesis to explain these results. Such a role for inflammation in resistance has been postulated, and some authorities consider it to be a highly effective mechanism in the body's defense against infection. Examples of similar phenomena have been reported. The lung of a dog which had recently recovered from a pneumococcal infection resolved an infection with an antigenically unrelated type of pneumococcus much more rapidly than a normal lung (Coggeshall and Robertson, 1935). More pertinent to the present study, mice have shown a markedly increased resistance to a

second infection with the cestode, *Hymenolepis nana* var. *fraterna*, as early as twelve hours after the first infection (Hearin, 1941), which is too brief a period of time for acquired resistance to be operative.

Both inflammation and tissue sensitivity have been suggested as possible factors in acquired resistance to animal parasites (Taliaferro, 1940). There is some evidence in the present results to support this hypothesis. Increased resistance to adult *T. spiralis* was demonstrated only when the concurrent agent, *A. caninum*, had passed through the small intestine a short time previously. As the time of this passage increased (from two to ten days), the effects on *T. spiralis* decreased. This would be expected if inflammatory factors were involved, as the tissue may undergo considerable repair within this period. Likewise, increased resistance to *T. spiralis* resulted in the lungs when *A. caninum* had occupied this organ previously causing striking damage. Finally, the effects of concurrent infection were similar in mice immunized against *T. spiralis* and in those infected with this parasite for the first time.

Because of the opportunities they afford to learn more about resistance to *T. spiralis* and other parasitic agents, it would seem desirable to continue studies of this type and to attempt to determine the mechanisms involved.

SUMMARY

Preliminary experiments are described in which information was obtained concerning the *A. caninum* infection in mice. This information was used in designing the final experiments testing the effect of an *A. caninum* infection on the natural and acquired resistance to *T. spiralis*.

In the four experiments on natural resistance, three groups of mice were used. The first and second groups, respectively, were infected orally and subcutaneously with 800 *A. caninum* larvae. The third group served as untreated controls. In the first two experiments, designed to test the effect of *A. caninum* on resistance to the adults of *T. spiralis*, one-half of the mice from each group were infected with *T. spiralis* two days and ten days, respectively, after the *A. caninum* infection. In the last two experiments, designed to test the effect of *A. caninum* on resistance to the larvae of *T. spiralis*, one-half of the mice of each group were infected with *T. spiralis* ten and twenty days, respectively, after the *A. caninum* infection.

In the first two experiments, all the mice infected orally with *A. caninum*, but not those infected subcutaneously, harbored significantly fewer adult *T. spiralis* than the controls. In the last two experiments, all the mice receiving *A. caninum* ten days prior to receiving *T. spiralis* harbored significantly fewer *T. spiralis* larvae than did the controls. In the twenty-day experiment, however, only the mice receiving *A. caninum* subcutaneously had significantly fewer *T. spiralis* larvae than the controls. These results show that in all cases, an oral or subcutaneous infection with *A. caninum*, prior to a *T. spiralis* infection, caused an increased resistance to *T. spiralis*.

In the two experiments on acquired resistance, two experimental groups and one control group of mice were immunized against *T. spiralis*. A fourth group

served as non-immunized controls. Two of the immunized groups were given 800 *A. caninum* orally or subcutaneously, while the third immunized group was given none. Two or ten days later, one-half of each group received a challenging infection with *T. spiralis*.

In both experiments (two and ten days), mice of the immunized control group harbored significantly fewer adult *T. spiralis* than the non-immunized controls, showing that acquired resistance to *T. spiralis* had been developed. Also, in both experiments, the immunized mice receiving oral *A. caninum* harbored significantly fewer adult *T. spiralis* than either the untreated, immunized controls, or the immunized group treated subcutaneously with *A. caninum*. The number of worms harbored by the latter two groups did not differ significantly. Therefore, acquired resistance to *T. spiralis* was increased in the mice treated orally with *A. caninum*, but was unaltered in those treated subcutaneously.

The mechanisms involved in the effects of *A. caninum* on resistance to *T. spiralis* were not studied, but the results show that cross immunity was not responsible. It is suggested that local inflammation resulting from the *A. caninum* infection was the important factor in producing increased resistance to *T. spiralis*.

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SEDIMENTARY VOLUMES OF THE COASTAL PLAIN SEDIMENTS OF NORTH CAROLINA

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Tulane University
New Orleans, Louisiana

The Coastal Plain of North Carolina covers 27,900 square miles of the State's total area of 51,900 square miles. The region has, within the last few years, been the scene of considerable geologic investigation for possible oil producing zones. Prospecting in these sediments, which are both marine and non-marine sands, clays and limes, has provided considerable data that give an excellent picture of subsurface conditions, as well as permitting a computation of their volume.

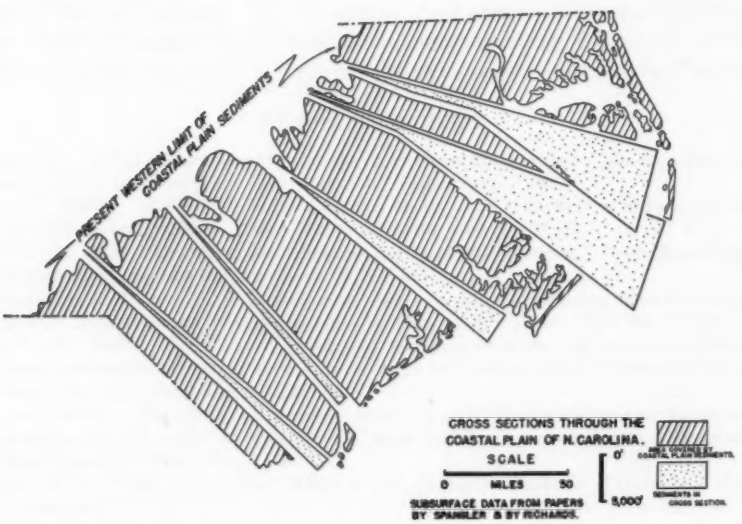
PRESENT WESTERN LIMIT OF COASTAL PLAIN SEDIMENTS
CROSS SECTIONS THROUGH THE COASTAL PLAIN OF N. CAROLINA.
SCALE
0 MILES 50
0'
5,000'
SUBSURFACE DATA FROM PAPERS BY SPANGLER & BY RICHARDS.

Fig. 1

The sedimentary accumulation today has thicknesses that vary from zero feet at the Fall Line, or contact between the older Piedmont or basement rocks and the Coastal Plain, to 1,200 feet in the area of the Great Carolina Ridge, and to more than 10,000 feet in the area of Cape Hatteras. By utilizing the depths to basement rocks, as given by Spangler (1950), Richards (1950), Straley and Richards (1948), and Skells (1950), a number of cross sections along the dip (Fig. 1) as well as along the strike (Fig. 2) have been drawn. These sections are based on proven depths to basement rock as obtained from drill data and not from estimation of geophysical data alone. An examination of the figures shows

that a large volume of sedimentary material remains in the Coastal Plain "wedge." By dividing the area into a number of small units and determining the average depths to basement rock in each of those units an approach to the total volume of materials can be made. This has been computed as being of the order of 11,700 cubic miles. On first consideration this appears as a large quantity of sediment by ordinary standards. Yet, when compared with the more than 1,000,000 cubic miles computed by Murray for the Gulf Coastal Plain, and reported in a personal communication, the North Carolina materials remain as only a small amount. These latter sediments go, with others, to constitute the Atlantic Coastal Plain. What additional figure must be added to arrive at the

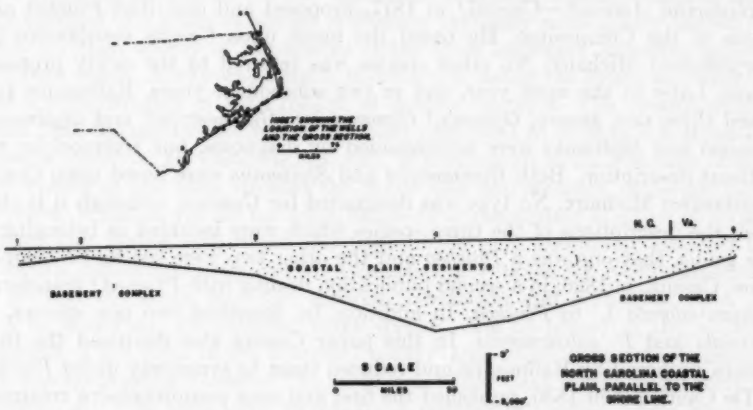

FIG. 2

volume of this sedimentory block is at present problematic but certainly not beyond computation.

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PLUCHEA, SECTION STYLIMNUS, IN NORTH AMERICA*

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Introduction

Plates 20–23

Historical Account.—Cassini,[1] in 1817, proposed and described *Pluchea* as a genus of the Compositae. He based the name upon *Conyza marilandica* (as *marylandica*) Michaux. No other species was referred to the newly proposed genus. Later in the same year, and in two subsequent years, Rafinesque proposed three new genera, *Gynema*,[2] *Gymnostyles* (*Gymnostylis*)[3] and *Stylimnus*.[4] *Gynema* and *Stylimnus* were accompanied by diagnoses, but *Gymnostyles* was without description. Both *Gymnostyles* and *Stylimnus* were based upon *Conyza marilandica* Michaux. No type was designated for *Gynema*, although it is clear from the descriptions of the three species which were included as belonging to the genus, that one was a *Pluchea* and the other two were species of *Gnaphalium*. Cassini, in 1826, in a second publication dealing with *Pluchea*,[5] transferred *Conyza odorata* L. to *Pluchea*. In addition, he described two new species, *P. petiolata* and *P. subdecurrens*. In this paper Cassini also discussed the three genera proposed by Rafinesque and reduced them to synonomy under *Pluchea*.

De Candolle,[6] in 1836, published the first and only comprehensive treatment of *Pluchea*. He transferred to the genus numerous species previously included in *Conyza*, *Baccharis*, *Erigeron*, *Eupatorium*, and the aforementioned genera of Rafinesque. This classical work has formed the basis for most subsequent treatments of the genus in various floras and manuals. With regard to some of the American species, De Candolle obviously understood imperfectly their delimitation and the type bases for their names. A state of taxonomic confusion was introduced by him, which in most cases has not been satisfactorily resolved to date. The object of the present study is to clarify the limits of the taxa, and the nomenclature pertaining to them, in the area under consideration.

Species of the genus *Pluchea* are widely distributed, chiefly within the tropics, and to a lesser extent in temperate regions. They occur in the Americas, in

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[1] H. Cassini. 1817. Bull. des Sci. Philomatique, p. 31.

[2] C. S. Rafinesque. 1817. Florula Ludoviciana, p. 63.

[3] ———. *Ibid.* p. 150; 1818, Am. Monthly Mag. Crit. Rev. **2**: 268.

[4] ———. 1819. Jour. Phys. Chim. Hist. Nat. **89**: 100.

[5] H. Cassini. 1826. Dictionnaire des Sci. Nat., Paris, **42**: 1.

[6] A. P. De Candolle. 1836. Prodromus **5**: 449–453.

Africa, and from Asia to Australia. *Pluchea* is one of a number of genera of the Compositae which have been assigned to the sub-tribe *Plucheineae* of the tribe *Inuleae*. Bentham[7] discussed at some length the problem as to whether some of these genera should be maintained as distinct from *Pluchea*, or included with it in a broad sense. If, in the geographical area under consideration here, an attempt were made to evaluate and redefine generic limits in this sub-tribe, it would require thorough study of the related Cuban genera, *Sachsia* and *Rodogeron*, and a genus of western North and South America, *Tessaria*. The status of several Old World genera similarly allied to *Pluchea* would require clarification, also.

The Scope of the Problem.—The taxonomic confusion which prompted this study pertains to a group of species of *Pluchea* obviously more closely related to each other than to any of the species of the three segregate American genera mentioned above. A way was sought, therefore, to define the group and thus to avoid a consideration of the problem of generic concepts. The present study reveals that the section *Stylimnus* has been employed to include the group of species in question. De Candolle (*op. cit.*), in treating all of the Plucheas of the world known at the time, divided the genus into two sections, *Stylimnus* and *Hebephora*. While he did not include a diagnosis or description of either of these, the American plants with which this paper deals were exclusively assigned to the section *Stylimnus*. Gray,[8] in his treatment of *Pluchea* for North America, also divided the genus into two sections, *Berthelotia* and *Stylimnus*, both of which he described. He included three species. One he placed in the section *Berthelotia*; it is a plant of the western United States which some authors assign to the segregate genus *Tessaria*. The other two, which Gray placed in the section *Stylimnus*, are here considered to involve a number of species. These species, misinterpreted by Gray, were, together with several others, similarly misinterpreted by De Candolle. Altogether, however, they form a natural group which both of these authors considered as comprising the section *Stylimnus*. This section name is employed here, therefore, in the sense of De Candolle's usage and as defined by Gray. The generic description in the systematic section below is purposefully limited to that section, and there is no attempt to include in the list of generic synonyms any other than those which pertain to it.

In the systematic treatment which follows, it will be noted that in the synonymy of certain of the species of *Pluchea* there are combinations of the specific epithets under *Placus*. *Placus* of Loureiro[9] is a synonymous earlier name for *Blumea*, a conserved Old World genus closely allied to *Pluchea* and one which might be included with it, if the latter were treated in an extended sense. Neither *Blumea* nor *Placus*, therefore, is identifiable with the section *Stylimnus* of *Pluchea*. The names are not included in the synonymy of this section of the genus, even though combinations of some of the species of *Pluchea* were made with *Placus*.

[7] George Bentham. 1873. Jour. of the Linnean Soc. (Bot.) **13**: 335–577.

[8] Asa Gray. 1884. Synoptical Flora of N. Amer. **2** (2): 225–226.

[9] Juan Loureiro. 1890. Flora Cochinchinensis **2**: 496–497.

Of the specimens studied, only a sufficiently large number were selected for citation to give both a true indication of the distribution of each taxon throughout its entire range, whether it were limited to the area under consideration or not, and a reasonable coverage of specimens from each lending institution. The herbaria from which specimens were borrowed for study, together with the symbols employed to indicate where those cited specimens are deposited, are as follows: University of California, Berkeley (C); Chicago Museum of Natural History, Chicago (CM); Duke University, Durham, N. C. (D); Botanical Museum, University of Copenhagen, Denmark (DN); University of Florida, Gainesville (F); Gray Herbarium, Harvard University, Cambridge, Mass. (G); University of Georgia, Athens (GA); Royal Botanic Gardens, Kew, Surry, England (K); National Arboretum Herbarium, Beltsville, Md. (NA); University of North Carolina, Chapel Hill (NC); North Carolina State College, Raleigh (NCS); New England Botanical Club Herbarium, Cambridge, Mass. (NE); New York Botanical Garden Herbarium, New York (NY); The Academy of Natural Sciences of Philadelphia (P); Southern Methodist University, Dallas, Texas (SM); University of Tennessee, Knoxville (T); United States National Herbarium, Smithsonian Institution, Washington, D. C. (US).

ACKNOWLEDGMENTS

The writer acknowledges his appreciation to the curators of the several herbaria from which specimens were made available for this study, and for the warm hospitality extended by the members of the staffs of the institutions where he worked during the course of the study. Grateful acknowledgment is made to the General Education Board, and to the American Association for the Advancement of Science through the North Carolina Academy of Science, both of which contributed to meeting the expense incurred. To Miss Marjorie Stone, Librarian of the Gray Herbarium, the writer is deeply grateful for her courtesy and helpfulness with bibliographic problems. Especially, thanks are due to Professor H. L. Blomquist, of Duke University, and Professor Lloyd H. Shinners, of Southern Methodist University, for their helpful advice and criticism.

GENERAL DISCUSSION OF NOMENCLATURAL PROBLEMS

The Confused Bases of the Names Pluchea odorata *and* P. purpurascens.—The bases for the names *Pluchea odorata* (L.) Cass. and P. *purpurascens* (Swartz) DC. require clarification. The first is a shrub the leaves of which are entire; the second is an herbaceous annual the leaves of which generally have crenate-serrate margins. Both occur in the West Indies, among other places, and it was from there that the plants were originally described.

The passages in the literature pertinent to an understanding of the bases of the original names, *Conyza odorata* and *C. purpurascens*, are given below; they are followed by a discussion leading to a clarification of their identities as presently understood.

The passage in Systema Naturae, ed. 10, 1213 (1759), in which Linnaeus described *Conyza odorata*, is as follows:

odorata. A. C. fol. ovatis serratis subtomentosis acutis, caule corymboso, coroll. subglobosis. *Sloan. jam.* t. 152. f. 1. Plum. ic. 97.

Later in the same year, in Amoentitates Academiae **5**: 406 (1759), Linnaeus listed *Conyza odorata*, without diagnosis, and gave the reference: "Brown. jam. 318."

The "Brown. jam." reference in the Amoen. Acad. is on page 318 of Patrick Browne's, "The Civil and Natural History of Jamaica" (1756), where the following account appears:

Conyza 1. Odorata minor erecta, purpurascens, corymbosa; foliis ovatis, villosis.

Conyza major odorata . . . . . Sloan. Cat. 124. h. t. 152. f. 1.

The smell of this plant is agreeable to most people: it is frequent in all low marshy lands, and seldom rises above sixteen or twenty inches in height. The stalk is generally pretty simple below the middle; but, as it rises, it throws out a good many branches, which reach nearly to an equal height, and carry their flowers in a shady spreading form at the top.

Swartz, Prodromus **112** (1788), described *Conyza purpurascens* as follows:

purpurascens—foliis ovato-lanceolatis serratis subtomentosis, caule subherbaceo, inferne simplici superne corymboso, floribus ovatis.
Conyza 1. Brown. Jam. 318.
Conyza major odorata . . . . Sloan. h. I. 258. t. 152. f. 1.
Jamaica.

The Sloane figure to which reference is made by both Browne and Linnaeus portrays a fragment of the uppermost portion of a plant with entire leaves. Except for the identity of this character, the figure is of little value in interpreting the plants in question. The Plumier plate, which is the second to which Linnaeus refers, consists of two parts. The principal figure portrays the entire uppermost portion of a plant with serrate leaves. This is sufficiently well done to identify the drawing with the herbaceous plant. The second figure on the Plumier plate is of a single, entire-margined leaf. It is unmistakably like the leaf of the shrubby plant, and it corresponds closely to the leaves shown in the Sloane figure.

An analysis of the Sloane and Plumier plates, correlated with the accounts of Linnaeus and Swartz, plainly lead to the conclusion that the bases for the two names were confused by the original authors. Linnaeus based *Conyza odorata* upon (1) Sloane's figure, and (2) Plumier's plate, which consisted in principal part of a figure unlike Sloane's, and of a single leaf comparable to the leaves of Sloane's figure. Linnaeus' diagnosis, moreover, must have been drawn from the principal figure of the Plumier plate. Swartz based *C. purpurascens* upon (1) Browne's description and discussion, and (2) the same Sloane figure. Fortunately,

the first element in the Linnean citation, the Sloane figure, can be related only to the entire-leaved, shrubby plant, while the first element in the Swartz citation is the Browne description and discussion which very clearly can be related only to an herbaceous plant. Urban, apparently quite aware of the confusion, resolved it in the case of *C. odorata*, for he typified the name as follows: "*Conyza odorata* L. Syst. X ed. II (1759) p. 1213 (quoad syn. Sl. et. ic. Plum. fol. separ.),"[10] and "Syst. X ed. II (1759) p. 1213 (excl. syn. Plum., quoad plant. foliis serratis)."[11]

Despite the original confused basis of the names, the Linnean name has come to be universally applied to the shrubby plant, and the Urban typification soundly establishes the nomenclature. Since the Swartz name has been similarly taken up by most authors for the herbaceous plant and is well understood, it is typified in the same fashion in the systematic treatment below.

The Identities of Pluchea camphorata *and* P. purpurascens—Fernald,[12] in 1939, satisfactorily settled the identity of *Pluchea camphorata* and clarified the basis for the name. That plant is the marsh fleabane (principally of fresh-water, rarely of brackish habitats) which was then being treated by authors as *P. petiolata* Cass. or *P. viscida* (Raf.) House. It occurs from Delaware to Florida and Texas, and in the interior northward to Missouri, Illinois, Indiana and Ohio. At the same time, Fernald indicated that the correct name for the so-called salt marsh fleabane (principally of salt, less frequently of fresh-water habitats) of the Gray's Manual range, treated in Gray's Manual, ed. 7 (1908), as *P. camphorata*, was *P. marilandica* (Michx.) Cass. In 1942, however, after further consideration, Fernald[13] recognized this salt marsh plant of the northeastern States as a varietal segregate of *P. purpurascens* (Sw.) DC. which occurs chiefly along the coast from Virginia to California, to some extent inland in the southwestern United States, in Mexico, Central America, northern South America, and in the West Indies. At that time, Fernald, recognizing this as one species complex, with the population of the northeastern United States varietally distinct, chose not to take up the later name *marilandica* for the variety. He could not locate the type specimen and hence could not establish the basis for the name. He therefore gave it the new name *P. purpurascens* var. *succulenta*. This was a correct taxonomic interpretation, and even though Fernald could not establish the basis for the name so that it could be taken up for the northeastern segregate, he was justified, by the International Rules, in giving it a new name as a variety.

Pluchea purpurascens was originally described (as *Conyza purpurascens*)[14] from the West Indies in 1788. Since that time, the various floras of West Indian areas, and also those of Mexico, Central America, and Venezuela, have used that name

[10] I. Urban. 1920. Fedde, Repertorium Specierum Novarum Regni Vegetabilis Beihefte **5**: 58.

[11] ———. 1920. Symbolae Antillanae **8**: 722.

[12] M. L. Fernald. 1939. Rhodora **41**: 459.

[13] ———. 1942. Rhodora **44**: 227.

[14] Olof Swartz. 1788. Nova Genera et Species Plantarum seu Prodromus Descriptionum Vegetabilum, p. 112.

for the marsh fleabane of those areas. In the southwestern United States, however, the same plant has always been treated in all local works of the area as *P. camphorata*. In the southeastern United States, Small and Chapman used both *P. camphorata* and *P. purpurascens* for salt marsh fleabanes and, as already indicated, in the seventh edition of Gray's Manual, as in the second edition of Britton and Brown's Illustrated Flora, the name used was *P. camphorata*. Fernald was the first to examine critically specimens both of the principally salt marsh plant from throughout its range and of that of principally fresh-water habitats, to distinguish clearly between them, and to establish the fact that the salt marsh plant is wide ranging with two infraspecific populations. Furthermore, he clarified the nomenclature pertaining to both, and in Gray's Manual, ed. 8 (1950), his interpretations are brought together.

The Identities of Pluchea foetida *and its Allies.*—The *Pluchea foetida* complex constitutes a series of species distinguished by their perennial habit and their sessile, characteristically oblong, truncate to auriculate-clasping leaves. The general range of occurrence of the complex of species is from southern New Jersey to Florida and eastern Texas, in Mexico, in the Bahama Islands and the Greater Antilles, and in Brazil. The Brazilian plants have never been identified specifically with those from other parts of the range. The present studies indicate that the species occurring there are distinct from those in the area under consideration here; as they are known by different names there is no necessity for comment here on the names applied to them. In the southeastern States, from South Carolina to Louisiana, Small (1933) recognizes four taxa: *P. foetida*, *P. tenuifolia* Small, *P. imbricata* (Kearney) Nash, and *P. longifolia* Nash. In all of the remainder of the range, only those plants from the Dominican Republic have been named other than *P. foetida* (or *P. bifrons* DC., a synonymous name). Urban[15] named a collection of Eggers from the Dominican Republic *P. Eggersii* and subsequently one other collection of a plant determined as that species was made there by Ekman.

In eastern North Carolina, field studies revealed the presence of two taxa in the *Pluchea foetida* complex. One grew in swamp forests, ditches, pond margins, and in depressions generally, all places which tend to be permanently wet. Plants of the other taxon grew principally in savannas, and pine flatwoods, habitats which are alternately very wet or very dry, according to the season, and which are subject to frequent fire. Occasionally they were found to occur in ditches and on pond margins, but always places apparently subject to marked fluctuation of the water table. Plants of the first sort had flowers with creamy-white corollas, heads about 8 mm. high, phyllaries obtuse to acute at the apex, and the plants were larger and more suffused with purple. Plants of the second sort had flowers with rose-colored corollas, heads about 5 mm. high, acuminate phyllaries, and the superficial color of the herbage of the plant, besides being less suffused with purple, had a light grayish cast (which is largely obscured in drying!).

None of Small's (1933) descriptions corresponded to the characters of the rose-

[15] I. Urban. 1903. Symbolae Antillanae **3**: 407.

flowered plant referred to above; neither did the original descriptions of the species segregated from *P. foetida* in the southeastern States, i.e., *P. imbricata*, *P. tenuifolia*, or *P. longifolia*. In the descriptions of none of these, incidentally, is there any mention of corolla color. Fernald, however, in Gray's Manual, ed. 8 (1950), says of *P. foetida*: "flowers creamy." Also, Urban (*op. cit.*) refers to the flower color of his new species, *P. Eggersii*, as follows: "*floribus ex sicco sordide flavidis*," and in comparing it to *P. bifrons* [*P. foetida*] says of the latter: "*floribus roseis*."

At this point in the study, a large series of specimens of the *Pluchea foetida* complex from throughout the range was assembled from several herbaria. An analysis of the material showed clearly that both the white-flowered and the aforementioned rose-flowered series were abundantly represented. It was apparent from the names on the labels that, aside from the previously mentioned species segregated from *P. foetida* in the southeastern States, no one had perceived the existence of the distinctive rose-flowered form amongst the material determined as *P. foetida*. Further study indicated these facts: (1) a distinct white-flowered taxon occurs from southern New Jersey to central peninsular Florida, westward to eastern Texas, with a station in southeastern Missouri, and with two stations in the Dominican Republic; (2) a varietal segregate of the above occurs from southern Georgia to central Florida; (3) a very distinct taxon, white-flowered and with much larger heads, occurs in central and northern Florida; (4) a rose-flowered series, confined largely to the outer half of the coastal plain from eastern North Carolina through Georgia, extends throughout Florida, occurs westward to eastern Texas, in San Luis Potosi and on the Yucatan Peninsula, Mexico, and in the Bahama Islands and Cuba. This series was divisible into two varietal tendencies. Number (2) above was the form called *Pluchea imbricata*; number (3) was *P. longifolia*; *P. tenuifolia* was a minor leaf form identifiable with number (1); and numbers (1) and (4) included the large mass of material passing as *P. foetida*, with the exception of that from the Dominican Republic which had been called *P. Eggersii*.

As between the white-flowered and rose-flowered forms, both of which have, in large part, been considered as *Pluchea foetida* (L.) DC., it is not difficult to establish which should be properly called *P. foetida*. *Baccharis foetida* L.,[16] upon which De Candolle based his *Pluchea foetida* combination,[17] rests upon a description of Gronovius.[18] This in turn was based upon a Clayton specimen, no. 159, from Virginia. Although the fact that there is no rose-flowered form from Virginia in the large series of specimens at hand is but negative evidence as to what Clayton had, Gronovius' (*ibid.*) phrase, "*Eupatorium floribus albus*," is sufficient to settle the identity even if the locality were not enough. In addition, a photograph of the Clayton specimen readily shows that it is unquestionably the white-flowered form. Interestingly, Walter,[19] commenting upon his *Baccharis viscosa*

[16] C. Linnaeus. 1753. Species Plantarum, p. 861.
[17] A. P. De Candolle. 1836. Prodromus 5: 452.
[18] J. F. Gronovius. 1739. Flora Virginica, p. 96.
[19] Thomas Walter. 1788. Flora Caroliniana, p. 202.

(a photograph of which in the Gray Herbarium definitely shows it to be of the *P. foetida* complex) says: "*Varietates, floribus albis, et floribus rubris.*" This is the only reference, in treatments of this complex of plants for the southeastern United States, in which "red" flower color is mentioned.

A specimen of Eggers, no. 2681, "Hab. in Sto. Domingo in Savanna de Cupey prope Loma Isabel de la torre 360 m. alt., in graminosis, m. Jun. flor.", upon which Urban based the name *Pluchea Eggersii*, was secured from the Kew Herbarium for study. The plant appears to be unquestionably identifiable with *P. foetida* (L.) DC. Furthermore, Urban's description and comments lead inescapably to the view that he was segregating an unusual white-flowered West Indian form from the more common rose-flowered form known to him as, and generally passing as, *P. bifrons* [*P. foetida*].

Since no name has been discovered to apply to any of the rose-flowered series, it remains to be described in the systematic treatment below.

SYSTEMATIC TREATMENT

BIBLIOGRAPHY OF *PLUCHEA*, SECTION STYLIMNUS

PLUCHEA Cassini, Bulletin des Sciences, par La Société Philomatique, de Paris 31 (1817); Dict. Sci. Nat. **42**: 1–2 (1826).

Gynema Raf., Florula Ludoviciana 63 (1817).

Gymnostyles Raf., *nomen nudum*, Florula Ludoviciana 150 (1817); not *Gymnostyles* Juss.

Gymnostylis Raf., *nomen nudum*, Am. Monthly Mag. Crit. Rev. **2**: 268 (1818).

Stylimnus Raf., Jour. Phys. Chim. Hist. Nat. **89**: 100 (1819).

Conyza subg. *Leptogyne* Ell., Sk. Bot. of S. C. and Ga. **2**: 322 (1824).

Pluchea sect. *Stylimnus* DC., Prodr. Syst. Nat. Regni Veg. **5**: 450 (1836); Gray, Syn. Fl. N. A. **1**(2): 225 (1884).

COMPOSITAE, tribe INULEAE, subtribe PLUCHEINEAE [Bentham and Hooker, Genera Plantarum **2**(1): 180 (1873)].

GENERIC DESCRIPTION

Heads with very numerous flowers, heterogamous. Central flowers of the head relatively few in number, bisexual, but sterile. Outer flowers of the head very much more numerous, in many series, pistillate and fertile. Receptacle flat, naked. Involucre imbricate, the phyllaries herbaceous to chartaceous.

Pappus of all flowers a single series of capillary, hispid bristles. Corollas rose to rose-purple, or creamy-white, in some species the inner bisexual flowers have purplish corollas while the outer pistillate have creamy ones. Corollas all tubular: those of the central bisexual flowers with the upper third portion dilated into a cylindrical or slightly campanulate tube with five deltoid lobes at the summit; those of the outer pistillate flowers filiform, very slightly dilated at the very summit which is divided into three slender lobes. Anthers (of the central bisexual flowers) connate, exserted from the corolla tube; each anther with a free, deltoid, apical appendage, and each having a deltoid, basal appendage which is connate with that of the contiguous anther and the whole basal appendage is prolonged into a filiform tail. Styles filiform: in some species the central bisexual flowers with an undivided stigma, in others with a bifid stigma the lobes of which are slender-elongate; those of the outer pistillate flowers always with a bifid stigma with slender-elongate lobes. Stigmas all flattened, the apices rounded, but with the margins much inrolled so that they seem, without careful examination, terete. The outer stigmatic surface is papillose throughout. Achenes

tiny, not exceeding 1 mm. in length, cylindrical, dark brown or reddish-brown in color, 4–6 angled, or with prominent, ridges and grooves, setose to hirtellous, or glabrous, and with a whitish caruncular base. Pappus of 1 series of barbellate bristles.

Heads numerous (rarely few), ovoid, or campanulate to hemispherical, disposed in panicled, elongate to flat-topped cymes.

Aromatic, annual or perennial herbs, or suffrutescent or frutescent plants of North and South America, Africa, and Asia to Australia. Stems from glabrous to glandular-pubescent to floccose, sometimes winged. Leaves alternate, simple; entire, crenate-serrate to serrate; ovate to linear-lanceolate; glabrous, or glandular-pubescent to floccose, petioled, sessile, auriculate-clasping, or decurrent.

ARTIFICIAL KEY TO THE TAXA

1. Plants definitely shrubby; leaves large, oval, oblong-elliptical, or elliptical, up to about 15 cm. long and about 6 cm. wide, silky-pubescent.....1. *P. odorata*
1. Plants annual or perennial herbs, or suffrutescent; if the latter, then the leaves lanceolate to linear-lanceolate, not silky-pubescent.
2. Leaves conspicuously decurrent, thus the stems winged.
3. Heads hemispherical, with broad, flat summits; phyllaries loosely imbricate in 2–3 series, scantily appressed-tomentose.....2. *P. suaveolens*
3. Heads narrowly to broadly campanulate, the summits convex; phyllaries imbricate in more than three series, glandular, or glandular and pilose to copiously but loosely tomentose.
4. Leaves lanceolate, gradually diminishing in size upwards on the upper stems and inflorescence branches from about 12 to 4 cm. in length; leaves from sharply and unevenly serrate-dentate to shallowly so, or essentially entire; heads *ca.* 5–7 mm. high and 5–7 mm. broad.
5. Peduncles glandular and sparsely pubescent with long, loosely spreading trichomes; the outermost phyllaries glandular and sparsely pubescent with loosely spreading trichomes.....3a. *P. salicifolia*, var. *salicifolia*.
5. Peduncles glandular, but the glands mostly obscured by a loose mat of woolly, white pubescence which at the summits of the peduncles tends to invest the bases of the involucres; outermost phyllaries copiously pubescent. 3b. *P. salicifolia*, var. *canescens*
4. Leaves lanceolate to linear-lanceolate, of nearly uniform size on the stems and inflorescence branches, up to about 5 cm. long, evenly and conspicuously dentate-serrulate; heads *ca.* 10 mm. high and 10 mm. broad..4. *P. parvifolia*
2. Leaves not decurrent, the stems not winged.
6. Leaves broadly elliptical to lanceolate or ovate to ovate-lanceolate, petioled or sessile, if sessile the bases of the blades narrowed to the mid-ribs, neither auriculate-clasping nor truncate.
7. Leaves petioled; inflorescence characteristically an elongate panicle; the branches numerous and terminating in convex, panicled cymes; the central uppermost axis maturing first, but the lateral ones never equalling or exceeding it; occasionally the inflorescence consisting of only a single, terminal, panicled cyme; phyllaries granular, only the outermost puberulent and ciliate.....5. *P. camphorata*
7. Leaves sessile or petioled; inflorescence characteristically more truly cymose, the younger lateral branches elongating and exceeding the more central ones, thus producing a flat-topped or layered inflorescence; phyllaries glandular or not, the outermost and median ones copiously puberulent and ciliate, the inner ones sparsely puberulent on their summits.
8. Heads 5–7 mm. high, 5–7 mm. broad; plants seldom less than 5 dm. tall, usually taller, up to 1.5 m.....6a. *P. purpurascens*, var. *purpurascens*
8. Heads 8–10 mm. high, 7–10 mm. broad; plants seldom more than 6 dm. tall, often much less.....6b. *P. purpurascens*, var. *succulenta*

6. Leaves oblong to oblong-ovate, or oblong-elliptical to oblong-lanceolate, sessile, the bases of the blades auriculate-clasping to truncate, not narrowed to the midribs,
9. Heads 10–12 mm. high; median phyllaries 1–2 times as long as broad; leaves conspicuously large, close, and overlapping, the median seldom less than 1 dm. long and 5 dm. broad, sharply, coarsely, and unevenly dentate-serrate to crenate-serrate ..... 7. *P. longifolia*
9. Heads 4–10 mm. high; median phyllaries 3–4 times as long as broad; leaves much smaller, the median seldom over 8 cm. long and 3–4 cm. broad, not conspicuously close and overlapping, serrate, dentate-serrate or serrulate, the teeth apiculate
10. Corollas creamy-white; heads 8–10 mm. high; outer phyllaries obtuse or obtuse-apiculate.
11. Heads about 8 mm. high, broadly campanulate; phyllaries loosely imbricate, green or stramineous ..... 8a. *P. foetida* var. *foetida*
11. Heads about 10 mm. high, turbinate-campanulate; phyllaries closely imbricate, the outer green, the inner rose-purplish ..... 8b. *P. foetida*, var. *imbricata*
10. Corollas rose-purplish; heads 4–6 mm. high; outer phyllaries acuminate.
12. Upper stems and inflorescence branches copiously pubescent; phyllaries glandular and copiously pubescent (at least on the upper portions) ..... 9a. *P. rosea*, var. *rosea*
12. Upper stems and inflorescence branches papillose-glandular with a slight tendency to a sparse pubescence; phyllaries papillose-glandular to sparsely pubescent ..... 9b. *P. rosea*, var. *mexicana*

TREATMENT OF THE SPECIES

1. *Pluchea odorata* (L.) Cass., Dict. Sci. Nat. **42**: 3 (1826)

Conyza odorata L., Syst. Nat. ed. 10, 1213 (1759). (As to the Sloane plate and single leaf of the Plum. ed. Burm. plate on which based, not as to plant described.)
C. carolinensis Jacq., Collectanea **2**: 271 (description) (1788); Ic. Pl. Rar. **3**: pl. 585 (1788).
C. Cortesii H. B. K., Nov. Gen. Sp. **4**: 75 (1820).
Pluchea Cortesii (H. B. K.) DC., Prodr. **5**: 452 (1836).
P. carolinensis (Jacq.) Sweet, Hort. Brit. ed. 3, 350 (1839).
Placus odoratus (L.) [incorrectly attributed to Baill. by] Gomez, Ann. Hist. Nat. Madrid **19**: 273 (1890).
Pluchea odorata (L.) Cass., var. *brevifolia* O. Kuntze, Rev. Gen. **1**: 357 (1891).
P. odorata (L.) Cass, var. *normalis* O. Kuntze, *ibid.*

Much-branched shrub, 3–4 (rarely 5) m. tall; lower stem glabrescent, bark finely ridged, smooth, grayish-brown or reddish-brown; upper stems and inflorescence branches cinereous or sordid-tomentose; leaves petiolate, the petioles 1–2.5 cm. long; leaf blades large, bright green above, pale below, oval to elliptical or oblong-ovate, 6–20 cm. long, 2.5–8 cm. broad, entire or very rarely repand-denticulate, or serrate-dentate sparsely short-pubescent above, except the younger which are copiously short-pubescent above; blades beneath silky, sub-tomentose to tomentose, pale or often cinereous or sordid, especially on the veins; inflorescences terminal, convex panicled cymes; heads broadly campanulate, *ca.* 6–7 mm. high, 6–8 mm. broad; outer and median phyllaries somewhat glandular, copiously pubescent, and the outer, especially, copiously long-ciliate, grading from broadly ovate or suborbicular and broadly obtuse on the outside of the involucres to oblong, broadly obtuse in the median portions, and linear-acute to attenuate on the inside, the innermost ones stramineous and essentially glabrous; corollas rose-purplish. FIG. 10, PLATE 22.

Distribution: Mexico, Central America, northern South America, and from Bermuda and the Bahama Islands through the West Indies to Trinidad; introduced in Southern Florida and the Hawaiian Islands.

Some representative specimens examined: **Mexico.** TAMAULIPAS: vicinity of Tampico, alt. about 15 m., *Palmer 75* (CM, G, NY, US); VERA CRUZ. banks of Río de los Pescados, Puente Nacional, *Purpus 12078* (G, NY); NUEVO LEÓN: moist soil near the stream, in gray silt between large pebbles, on an island in Río Santa Catarina about a mile above the entrance to Cañón de Huasteca, about 5 miles from Villa Santa Catarina, *Hernández et al. 44M801* (G); HIDALGO: rocky mountainside, alt. 4500 ft., Jacala, *Chase 7168* (CM, G, NY); PUEBLA: near Metlaltoyuca, *Goldman 71* (G, US); GULF OF CALIFORNIA. SAN MARCOS ISLAND: about pools in ravines cut into gypsum, forming rank growths 4–15 ft. high, *Johnston 3632* (C, G, NY, US); BAJA CALIFORNIA: riparian under trees in canyon, a heavy-stemmed, polypodial, leafy bush, 2–3 m. tall, Rn. Primera Agua, Sierra Giganta, *Gentry 3717* (C, G); SINALOA: Mazatlán, *Ortega 5700* (US); NAYARIT: San Blas, Tepic, *Maltby 24* (US); MARIA MADRE ISLAND: Arroya Honda, shrub 10 ft. high, in stream beds, *Mason 1773* (CM, G); JALISCO: Ixtapa, alt. 200–300 ft., *Nelson 4144* (G, US); COLIMA: western edge of San Pedrito Lagoon, vicinity of Manzanillo, *Ferris 6132* (US); MICHOACÁN: roadside, silty-sandy soil, shrub up to 10 ft. high, 0.6 mile n. of Coahuayana, 20 miles s.e. of Tecoman, *Turner 1991* (SM); CHIAPAS:? Guxbla, Cuesta unterh. Hac. Petapa, guellige Abhange, Feb. 18, 1896, *Seler 1910* (G, NY); OAXACA: Etla, alt. 3500 ft., *Smith 369* (G); CAMPECHE: Canasayab, *Lundell 1418* (CM, NY); YUCATAN: Izamal, *Gaumer 399* (CM, DN, G, NY, US).

British Honduras: tall shrub growing in open forest, flowers pale puce, leaves when crushed fragrant, "rare," All Pines, *Schipp 762* (C, CM, G, NY); **Guatemala:** wet thicket, wet forest near Tactic, above the bridge across Río Frío, alt. 1400–1500 m., Alto Verapaz, *Standley 90391* (CM). **Honduras:** moist thicket along stream, vicinity of Comayagua, about 600 m., Comayagua, *Standley & Chacon P. 5894* (CM). **El Salvador:** shrub along stream, near Colon, La Libertad, *Williams & Molina R. 15205* (CM). **Nicaragua:** environs de Grenade, forest *Levy 426* (DN). **Panama:** "Salvia" bought of street vendor. Tea made of leaves used for sore throat, *Macbride 2766* (CM, US). **Colombia:** Barranquilla and vicinity, *Bro. Elias 685* (US). **Venezuela:** hills of Los Mariches and vicinity of Petare and El Encantado, in forests and savannas, alt. 500–1000 m., Miranda, *Pittier 9269* (G, US). **Brazil:** (no locality) *Lund* (DN). **Ecuador:** (no locality) *Eggers 14853* (CM).

Bermuda: hillside, Paynter's Vale, *Brown & Britton 262* (CM, G, US, NY). **Bahama Islands:** clearing, 8-mile rocks, Great Bahama, *Britton & Millspaugh 2575* (CM, NY). **Cuba:** limestone plain, Boquerón, Ensenada de Siguanea, Isle of Pines, *Britton et al. 14530* (NY); along creek at Cruceros, vicinity of Soledad, Las Villas, *Howard 4857* (G, NY); bed of river, Valley of the Rio Bayamita, south slope of the Sierra Maestra, alt. 900–1050 m., Oriente, *Maxon 3917* (CM, NY, US). **Jamaica:** along road, Orange River Valley, near Montego Bay, *Maxon & Killip 1683* (CM, G, US). **Haiti:** river bank near mouth of Trois Rivières, vicinity of Port de Paix, *Leonard & Leonard 11765* (G, US). **Dominican Republic:** Paradis prope Barahona ad Rio Nizaito, Barahona, *Turckheim 2809* (CM, G, NY, US). **Puerto Rico:** in mangrove swamp, near Martin Peña Station, *Heller 4873* (CM, G, NY, US). **St. Croix:** Catherine's Rest, *Ricksecker 276* (C, CM, G, NY, US). **Guadeloupe:** *Questel 644* (US). **St. Vincent:** hillside plant, Silver Spoon District above Three Rivers, *Howard 11187* (G, NCS). **Trinidad:** Tabaquite, Sept. 13, 1918, *Broadway* (G, NY, US). **United States, Florida:** in dry sandy soil along roadside, Cocoanut Grove, Dade Co., *Moldenke 731* (D, NY). **Hawaii:** between Barber's Point and Pearl Harbor along beach, naturalized and rapidly spreading, Oahu, May 8, 1932, *Degener 18079* (G).

Upon examination of the specimens of *Pluchea odorata* from throughout its range a number of specimens from Andean regions of southern Bolivia and northwestern Argentina were encountered, amongst the material from the several

lending herbaria, which had been determined as *P. odorata*. Among them was the type specimen of "*Pluchea odorata* ? var. *ferruginea* Rusby." However, the Andean plant, a shrub which superficially resembles *P. odorata*, is quite distinct from that species. It is *P. fastigiata* Grisebach; a number of other specimens of it from the same region, correctly determined, were examined also. *P. odorata* is not known from any place in western South America to the south of Ecuador.

2. *Pluchea suaveolens* (Vell.) O. Kuntze, Rev. Gen. Pl. **3**(2): 168 (1898)

Gnaphalium suaveolens Vellozo, Flora Fluminensis **8**: tab. 100 (1827).
Pluchea Quitoc DC., Prodr. **5**: 450 (1836).

Herbaceous perennial, 0.5–2 m. high, usually with several erect branches; stems and leaves (both sides) glandular and appressed-pubescent, glabrate below, the younger leaves and upper stems tomentose; stems winged by the decurrent leaf bases; leaves sessile, decurrent, lanceolate to broadly lanceolate, rarely linear-lanceolate on the one hand, or ovate on the other, shallowly serrate or serrate-crenate; median leaves 8–14 cm. long and 2–4 cm. broad; inflorescence cymose-paniculate, the heads on short or relatively long, lax peduncles; heads hemispherical with broad, conspicuously flat summits, *ca.* 6 mm. high and 10–12 mm. broad; phyllaries loosely imbricate in 2–4 series, the outer oval-oblong, short acuminate, herbaceous, grading to linear-attenuate and stramineous on the inside; corollas white or rose-purplish. FIG. 14, PLATE 22.

Distribution: Moist, open habitats in Argentina, Uruguay, Paraguay, Bolivia and Brazil; an adventive weed of moist coastal areas of Florida and Alabama, and presumably adventive in Trinidad.

The basis for the name *Gnaphalium suaveolens* is a figure of the plant, tab. 100 in Vellozo, Flora Fluminensis.

Some specimens examined: **Argentina.** BUENOS AIRES: Punta Lara, La Plata, *Krapovickas 2894* (US); CÓRDOBA: Dec. 1891, *Kuntze* (NY); CHACO: Margarita Belen, *Myer 35757* (G, NY); CORRIENTES: Lomas Carambola, *Ybarrola 396* (G); TUCUMAN: Dep. Cruz Alta, *Ousset 134* (G, NY); Dep. Burroyaco, Cesso del Nogalito, *Venturi 8813* (G, US); FORMOSA: *Jorgensen 2751* (G, US); SALTA: Tortagal, *Parodi 9246* (G). **Uruguay.** MONTEVIDEO: Cerro, Lit. Platens, *Herter 289* (C, CM, NY, G); SAN JOSÉ: Barra de Sta. Lucía, *Herter f. 280A* (NY, US); SORIANO: Juan Jackson, *Monzón-Heber PE4750* (G). **Paraguay:** in regione lacus Ypacaray, *Hassler 11712* (DN, G, US); Central P., *Morong 619* (C, CM, G, NY, US); Cordillera de Villa Rica, *Hassler 8836* (G). **Bolivia:** sandy loam, Cururenda, Chaco, *Cardenas 2517* (US); Buena Vista, Santa Cruz, *Steinbach 5272* (CM, NY, G); Río Palometillas, Santa Cruz, *Steinbach 6762* (CM, G); Chaco-Andinis, *Fries 1075* (US). **Brazil.** RIO GRANDE DO SUL: Canoas, *Luis 29* (CM, US); "SUDBRASILEIN": *Bornmüller 638* (G); SAÕ PAULO: Belenziuho, *Hoehne 17115* (US); Arbusto, S. Luiz Parahytinga, *Viegas 3483* (US); RIO DE JANERIO: Wilkes Exp. 1838–42 (G, US); clay bank, Avenida Niemeyer, *Smith 2177* (G); open woods, *Holway and Holway 1292* (US); ALAGOAS: *Gardner 1347* (CM). **Trinidad:** in damp land near the sea, Woodbrook, *Broadway 7490* (US); *Botanic Gardens Herb. 2046* (US). **United States. Florida:** miry ground near Pensacola, *Curtiss 6873* (C, G, NY, US); ballast wharf, Pensacola, *Curtiss 1377* (CM, NY, US); **Alabama:** ballast, Mobile River, July 20, 1891, *Mohr* (US); Hunters Wharf, ballast, Sept. 27, 1894, *Mohr* (US); Mobile, July 1896, *Mohr* (US).

Pluchea suaveolens occurs in the area under consideration here only as an adventive in Florida and Alabama. Moreover, all of the specimens of this plant

from these states were collected on ballast approximately half a century ago. In his "Plant Life of Alabama",[20] Mohr reported *P. adnata* (H. & B.) Mohr [P. salicifolia (Mill.) Blake], and Small (1933) lists *P. adnata* as having been "reported as adventive on ballast in Ala." Mohr's specimens, however, prove to be characteristic *P. suaveolens*. Even though there are no relatively recently collected specimens of this species from Florida and Alabama, there is no positive evidence that the plant no longer persists there. The species is, therefore, included here and it is hoped that botanists doing field work in Florida and Alabama will thus be encouraged to ascertain whether it has indeed persisted there to the present.

Inasmuch as it was intended that representative specimens of each species treated in this paper from throughout the entire range of occurrence of each be cited, all of the South American material from each lending institution was examined. Amongst this material were two specimens from northwestern Argentina determined as *P. suaveolens*, which, in habit and vegatative characters, superficially resembled that species. However, the specimens exhibit several important characters clearly distinguishing them from *P. suaveolens*. In addition, two other similar specimens, determined only to the genus, were encountered, one from the same area in northwestern Argentina, and one from Peru. These four specimens clearly represent a species distinct from *P. suaveolens*. It is, so far as can be determined, a species new to science. At the end of this paper, under the heading, "A New South American Species," it is named and described as *Pluchea microcephala* Godfrey.

3a. *Pluchea salicifolia* (Mill.) Blake, var. *salicifolia*, Contr. U. S. Nat. Herb. **26**: 237 (1930)

Conyza salicifolia Miller, Gard. Dict. ed. 8, *Conyza* No. 6 (1768).
Baccharis adnata Humb. & Bonpl. ex Willd., Enum. Plant. Hort. Berol. 870 (1809).
Conyza adnata (H. & B.) H. B. K., Nov. Gen. & Sp. **4**: 74 (1820).
Pluchea subdecurrens Cass., Dict. Sci. Nat. **42**: 4 (1826).
P. adnata (H. & B.) Mohr, Contr. U. S. Nat. Herb. **6**: 790 (1901).

Suffrutescent, about 1–2 m. high; stems glandular, sparsely pilose throughout or occasionally canescent below, with a dense tuft of pilosity in the leaf axils; inflorescence branches glandular and sparingly pilose, not canescent; leaves conspicuously decurrent, sessile, lanceolate to linear-lanceolate, acute, from sharply and unevenly serrate to shallowly and unevenly serrate-dentate, or essentially entire; leaves gradually diminishing in size upwards on the stem, on the upper three decimeters of the stem (all that was available for study) the larger about 12 cm. long and 2 cm. broad, often all much smaller; leaves papillose-glandular above and below, otherwise the upper surfaces of the older leaves glabrate to sparsely pubescent, their lower surfaces sparsely pilose especially on the veins, the younger leaves sparingly pubescent above and from sparsely pilose to copiously hirtellous and sparsely pilose to subtomentose below, the youngest leaves terminating the branchlets sometimes canescent below; inflorescence a panicled cyme, the heads on short peduncles; heads campanulate, 4–5 mm. high and 4–5 mm. broad; phyllaries loosely imbricate in several series, the outer and median glandular and pilose, the inner glabrous; outermost phyllaries

[20] C. Mohr. 1901. Contr. U. S. Nat. Herb. **6**: 790.

lanceolate-acute or acuminate grading inwardly to linear-attenuate, the tips often suffused with purple; corollas roseate, or white, or the outer filiform corollas of the pistillate flowers white or pale rose and those of the inner bisexual flowers more deeply roseate. FIG. 15, PLATE 22.

Distribution: Riparian and on dry slopes, Mexico and Guatemala.

Specimens examined: **Mexico**: Photograph of the type of *Conyza adnata* (US); TAMAULIPAS: *Runyan & Tharp 4068* (US); VERA CRUZ: on rocks in the River, Sulphur Spring, Zacuapán, Jan. 1907, *Purpus* (C); Panoaya, *Purpus 8996* (CM, US); SAN LUIS POTOSÍ: Guascama, *Purpus 5129* (C); PUEBLA: vicinity of San Luis Tultitlanapa, *Purpus 3121* (C, CM, NY, US); SONORA: canyon in Sierra de Alamas, vicinity of Álamos, *Rose et al. 12982* (US, NY); SINALOA: Rancho de los Chinacates, San Ignacio, alt. 100 m., *Montes and Salazar 163* (US); MICHOACÁN: Zamora, *Nelson 6542* (G, NY, US); MICHOACÁN ET GUERRERO: Arroyo de Parabas, *Langlassé 26* (US); STATE OF MEXICO: by the river, 1340 m. alt., Tejupilco, Temascaltepec, *Hinton 3025* (G); MORELOS: Valle de Cuernavaca, *Lyonnet 388* (US); OAXACA: Nochistlán, alt. 4500 ft., *Conzatti 142* (G). **Guatemala**: opp. El Rancho, alt. 2000 ft., Tierra de los Minas, Baja Merapaz, *Kellerman 4997* (US).

3b. *Pluchea salicifolia* (Mill.) Blake, var. *canescens* (A. Gray) Blake, Jour. Wash. Acad. Sci. **21**: 328 (1931)

P. subdecurrens Cass., var. *canescens* A. Gray, Proc. Amer. Acad. **5**: 182 (1861).
P. adnata (H. & B.) Mohr, var. *canescens* (A. Gray) Blake in Standley, Contr. U. S. Nat. Herb. **23**: 1510 (1926).

Like var. *salicifolia* but the stems, leaves, and inflorescence branches canescent and much less glandular; canescence to a considerable extent deciduous on the older portions of the stem and on the older leaves, persistent, however, on the inflorescence branches and the outer phyllaries; outer phyllaries more copiously pilose; heads larger, 6–7 mm. high and 6–7 mm. broad. FIG. 12, PLATE 22.

Distribution: Riparian and on dry slopes, Mexico.

Type Specimen: Wartenberg, near Tantoyuca, prov. Huasteca, [Veracruz], Mexico, 1858, *L. C. Ervendberg 343* (G).

Additional specimens examined: TAMAULIPAS: vicinity of Victoria, *Palmer 12* (C, CM, G, NY, US); 9 kilo. east of Palmillas on small creek, surrounding hills (limestone) dry, el. 1750 m., *Stanford et al. 944* (C, G); Jaumave, *Rozynski 33f* (CM); NUEVO LEÓN: Monterey, *Palmer 546* (G, US); streams, Monterey, 1600 ft., *Pringle 10198* (C, CM, DN, G, NY, US); JALISCO: bushy streamside (fl. pink), Las Mesitas, Sierra Madre Occidental, n.w. of San Sebastián, *Mexia 1878* (C, CM, G, NY, US); COLIMA: *Palmer 1097* (G, NY, US); MICHOACÁN: Coalcomán, 1000 m., (fl. white), *Hinton et al. 12961* (G, NY, US); Hacienda Coahuayula, *Emrick 133* (CM); MORELOS: Cuernavaca, *Deam 6* (G); GUERRERO: Acapulco and vicinity, *Palmer 441* (C, CM, G, NY, US).

The two varieties of *Pluchea salicifolia* are distinguished only by the degree of difference in glandularness and pubescence on the leaves, stems, peduncles, and involucres. Their general ranges of occurrence are essentially identical geographically. Data on labels of specimens is, however, not sufficient to indicate whether or not habitat differences or differences in time of flowering may exist. Lacking field experience with these plants, and therefore even a minimum knowledge of criteria often important in understanding the delimitations of in-

fraspecific categories in such populations, it is considered prudent to maintain the varietal segregates.

4. **Pluchea parvifolia** (A. Gray) Godfrey, comb. nov.

P. subdecurrens Cass., (DC. in error), ? var. *parvifolia* A. Gray, Proc. Amer. Acad. **5**: 160 (1861).

P. adnata (H. & B.) Mohr, var. *parvifolia* (A. Gray) Blake in Standley, Contr. U. S. Nat. Herb. **23**: 1510 (1926).

P. salicifolia (Mill.) Blake, var. *parvifolia* (A. Gray) Blake, Jour. Wash. Acad. Sci. **21**: 328 (1931).

Suffrutescent, about 1–3 m. high; stems and leaves copiously papillose-glandular and stipitate-glandular, sparsely pilose, the axillary branchlets copiously pilose at the nodes where they arise; leaves conspicuously decurrent, sessile, lanceolate to narrowly linear-lanceolate, acute to acuminate, evenly dentate-serrulate or apiculate-serrulate, tending to be uniform in size along the stem, up to about 7 cm. long and 1.5 cm. broad although often much smaller; inflorescence a panicled cyme, the heads on relatively long peduncles; heads broadly campanulate, about 10 mm. high and 10 mm. broad; phyllaries loosely imbricate in several series, copiously glandular, the margins with very few irregularly disposed cilia; outermost phyllaries lanceolate-acuminate grading inwardly to linear-attenuate, all with the tips suffused with purple; filiform corollas of the outer pistillate flowers whitish to faintly rose-purplish, those of the inner bisexual flowers conspicuously rose-purplish. FIG. 13, PLATE 22.

Distribution: Riparian, in southern Baja California.

Type Collection: Cape St. Lucas, etc., Lower California, Aug. 1859–Jan. 1860, *Xantus 53* (G, NY, US). When Gray described this plant, he was enumerating a collection of plants of Xantus "communicated to the Smithsonian Institution," hence the specimen at the National Herbarium is here considered the Type.

Some additional specimens examined: BAJA CALIFORNIA: along stream, Comondu, *Wiggins 5503* (C, G, NY, US); San Gregorio, Feb. 8, 1889, *Brandegee* (C, US); sides and bottoms of wash, 22 mi. s. of La Paz, *Wiggins 5606* (G, NY); San José del Cabo, 1899, *Grabendorfer* (C); San José del Cabo, *Purpus 309* (C, US); San José del Cabo, *Anthony 430* (US); hillside near San Antonio, *Shreve 7298* (C); riparian in moist granitic sands, shrubby perennial, Las Cuevas, Cape District, *Wiggins 4343* (C, G).

When Gray described this plant as a variety of *P. subdecurrens* [*P. salicifolia*], he was in evident doubt as to its affinity, as judged by the way he wrote the name, *viz*, "*Pluchea subdecurrens* DC.? var. *parvifolia*." He had but small, fragmentary specimens with immature heads. There is now available a sufficient number of well preserved, adequate specimens, both of this plant from its type locality, and of *P. salicifolia* (*sensu lat.*), to yield a satisfactory concept of each in relation to the other. The plant hitherto regarded as var. *parvifolia* of *P. salicifolia* is well isolated geographically. It occurs only in southern Baja California whereas *P. salicifolia* var. *salicifolia* and *P. salicifolia* var. *canescens* are of relatively wide occurrence in other parts of Mexico, but absent in Baja California. The differences in foliar characters and head size, as given in the key and de-

scriptions, are marked and conspicuous. The three taxa are obviously closely related; however, the two varieties of *P. salicifolia* are so much more nearly like each other than the Baja Californian plant is like either of them, that the latter is regarded as specifically distinct.

5. *Pluchea camphorata* (L.) DC., Prodr. **5**: 452 (1836)

Erigeron camphoratum L., Sp. Pl. 864 (1753).
Baccharis foetida sensu Walt., Fl. Carol. 202 (1788); not Linnaeus.
Conyza camphorata (L.) Pursh, Fl. Am. Sept. **2**: 523 (1814); Elliott, Sk. Fl. S. C. & Ga. **2**: 321 (1824).
Gynema dentata Raf., Ann. Nat. Ann. Synop. **15** (1820).
G. viscida Raf., *ibid.* (renaming of *G. dentata*).
Pluchea petiolata Cass., Dict. Sci. Nat. **42**: 2 (1826).
P. foetida sensu DC., Prodr. **5**: 452 (1836).
P. viscida (Raf.) House, Am. Mid. Nat. **7**: 129 (1921).

Annual or perennial herb, up to about 1.5 m. tall; stems glabrate below, puberulent on the upper portions and on the inflorescence branches, leafy to the summit; leaves petiolate, the petioles 1–2 cm. long; leaf blades elliptical to oblong-elliptical, *ca.* 6–15 cm. long, 3–7 cm. broad, the margins dentate-serrate to repand-serrate, or essentially entire; upper and lower surfaces of the blades with sparsely distributed resinous atoms, the lower surfaces puberulent; inflorescence characteristically an elongate panicle, the branches numerous and terminating in convex, panicled cymes; the central uppermost axis maturing first, but the lateral ones not nearly equaling or exceeding it; occasionally the inflorescence consisting of only a single terminal, convex, panicled cyme; heads campanulate, *ca.* 5 mm. high, 3–4 mm. broad; phyllaries granular with resinous atoms, the outermost sparingly pubescent and ciliate, the median and inner not pubescent, grading from ovate-obtuse on the outside of the involucres to oblong-obtuse in the median portions to linear-oblong and obtuse to acute on the inside; phyllaries all thinnish, the outermost herbaceous, the median and inner scarious and usually purplish or rose-colored; corollas rose-purplish. FIG. 17, PLATE 22.

Distribution: Fresh habitats: wet woodlands, marshes, ditches, swales, and shores, Maryland and Virginia southward to northern Florida, westward to eastern Texas and Oklahoma, northward to southern Illinois, Indiana, and Ohio.

Type Specimen: Clayton no. 165 from Virginia (in the Herbarium of the British Museum of Natural History). Fernald published a photograph of the type in Rhodora **41**: pl. 569 opp. p. 458 (1939).

Some additional specimens examined. **Delaware.** SUSSEX CO.: Sept. 9, 1875, *Commons* (NY). **Maryland.** TALBOT CO.: in damp woods. Easton, *Clark 8* (G). **District of Columbia:** Zoological Park, Washington, Sept. 1897, *Hunter* (US). **Virginia.** BEDFORD CO.: Sept. 15, 1871, *Curtiss* (G); CAROLINE CO.: swale south of Milford, *Fernald & Long 7689* (G); SOUTHAMPTON CO.: wet siliceous and argillaceous drained border of Predler's Pond, Nottoway Swamp, southwest of Sedley, *Fernald & Long 7690* (G). **North Carolina.** CABARRUS CO.: cart road, bank of Coddle Creek, w. of Concord, *Fox 5438* (F, G, NA, NCS, NY, SM); CHEROKEE CO.: September 1893, *Ashe* (NC); COLUMBUS CO.: edge of swamp, n. shore of Lake Waccamaw, *Fox & Whitford 1876* (NCS); DURHAM CO.: wet pasture, Duke Forest, Mud Creek below Cornwallis Rd., *Oosting 3319* (D); HARNETT CO.: swampy roadside, Erwin, *Oosting & Blomquist 33621* (D); PASQUOTANK CO.: Dismal Swamp, *Harris C17731* (G); PERSON CO.: Loch Lily,

Roxboro, *Blomquist 5298* (D, G); POLK CO.: Columbus, Oct. 1897, *Townsend* (US); ROWAN CO.: vicinity of Heilig's Mill P. O., *Small & Heller 91* (CM, NY); STOKES CO.: in opening in alder swamp, near U. S. Rt. 311, w. of Dan River, 3.5 miles e. of Walnut Cove, *Fox 5490* (D, DN, F, G, K, NA, NCS, NY). **South Carolina.** ABBEVILLE CO.: Abbeville Distr., Aug. 1855, *ex herb. Hexamer & Maier* (G); BEAUFORT CO.: small, low, wet opening in live-oak forest, Hunting Island, *Godfrey 50927* (D, NCS); BERKELEY CO.: clearing along logging railroad, floodplain forest, along the Santee River, 3 miles northeast of Pineville, *Godfrey 8152* (D, G, NY, US); OCONEE CO.: *Anderson 1599* (US). **Georgia.** BARTOW CO.: low area in woods in dry drainage exit from Quicksand Pond, 4.8 miles s.e. of Adairsville, *Duncan 13146* (G, NCS); BIBB CO.: low meadows on Ockmulgee River, *Smith 909* (GA, US); CHATOOGA CO.: bottomland pasture at Knox Spring, 4.5 miles n.e. of Meno, *Duncan & Venard 13092* (GA, NCS); LOWNDES CO.: edge of cypress pond, 5 miles e. of Lake Park, *Quarterman 466* (D); MILLER CO.: Spring Creek Swamp, Colquitt, *Thorne 6369* (G, GA); SCREVEN CO.: edge of limesink, *Eyles 7292* (D). **Florida.** ALACHUA CO.: hammock, low ground, Gainesville, Oct. 2, 1940, *Watson & Murrill* (F); COLUMBIA CO.: Lake City, *Nash 2484* (NY, US); HAMILTON CO.: Alapaha River bank, n.w. of Jasper, Sept. 30, 1941, *West & Arnold* (F); JACKSON CO.: edge of bog, 8 miles e. of Marianna, *Hood 2666* (F).

Ohio. HAMILTON CO.: Cincinnati, *Turner* (G). **Indiana and Illinois.** LOWER WABASH VALLEY: Fordyce bottom, *Shearer 10302* (D). **Indiana.** CLARKE CO.: abundant in a drained swamp in a woods about 2 miles n. of Jeffersonville, *Deam 24387* (NY); HARLAN CO.: near Poor Fork P. O., *Kearney 272* (G, US); JACKSON CO.: plentiful along the right-of-way of the traction line, about ½ mile s. of Chestnut Ridge, *Deam 14041* (NY, US); JEFFERSON CO.: Hanover, Sept. 1875, *Young* (C, NY); POSEY CO.: low, clay woods, Erwin Woods, 5 miles s.w. of Hovey, *Tryon 4254* (CM, G, US). **Illinois.** ALEXANDER CO.: swampy ground, 6 miles n. Thebes, *Winterringer 5445* (CM); PULASKI CO.: swamps and sloughs, *Winterringer 2510* (CM); RICHLAND CO.: lower Wabash Valley, *Ridgway 2859* (CM); WABASH CO.: vicinity of Mt. Carmel, Sept. 1877, *Patterson* (CM). **Kentucky.** HARLAN CO.: Poor Fork P. O., *Kearney 272* (CM, NY); LAUREL CO.: swamps, e. of London, Sept. 5, 1934, *Braun* (NY); LYON CO.: Kuttawa, *Eggleston 5232* (NY); MCCRACKEN CO.: bottomland woods along Island Creek, 3 miles s. of Paducah, *Duncan 6109* (GA). **Tennessee.** BENTON CO.: swamp, west of Camden, *Sharp et al 5934* (NCS, T); BLOUNT CO.: creek bank, Tab Cat Creek, Great Smoky Mts. National Park, *Sharp 811* (T); COFFEE CO.: wet places, Tullahoma, *Biltmore Herbarium 4377b* (G, NY, US); DAVIDSON CO.: near Nashville, Aug. 30, 1877, *Ward* (US); LAUDERDALE CO.: Hatchie River bottoms s. of Ripley, *Shanks et al. 13571* (NCS, T); LINCOLN CO.: low ground, Elora, *Biltmore Herbarium 4377d* (US); MONTGOMERY CO.: along Dover road west of Datsonville, *Shanks 14410* (T); TIPTON CO.: old field, e. of Covington, *Sharp 6430* (T); WAYNE CO.: in gravel along a stream, 6 miles w. of Waynesboro, *Svenson 4317* (G.). **Alabama.** CLAY CO.: *Earle 907* (NY); HENRY CO.: thickety low stream bank, in rich sandy soil, 8 miles north of Headland, *Wiegand & Manning 3292* (G); LEE CO.: Auburn, *Earle & Baker 272* (NY); MOBILE CO.: sandy ditch, flat pine land, 11.5 miles s. of Mobile, *Shinners 12747* (SM); ST. CLAIR CO.: Asheville, *Vasey* (CM, NY). **Mississippi.** ATTALA CO.: McCool, Sept. 19, 1891, *Seymour* (D); CHOCTAW CO.: Ackerman, Sept. 19, 1891, *Seymour* (D); GRENADA CO.: Grenada, Sept. 25, 1891, *Seymour* (D); LEAKE CO.: creek bottom, Natchez Trace Parkway, *McDougall 1141* (US); LEE CO.: Tupelo, Sept. 22, 1891, *Seymour* (D); MUNROE CO.: Aberdeen, Sept. 21, 1891, *Seymour* (D); PANOLA CO.: clay soil, 10 miles w. of Sardis, *Broadhead 96* (SM); UNION CO.: New Albany, Sept. 23, 1891, *Seymour* (D). **Missouri.** BARRY CO.: Eagle Rock, *Bush 71* (NY, US); BUTLER CO.: Poplar Bluff, Aug. 1879, *Letterman* (US); upland pond along road, 2 mi. s.w. of Wappapello, *Steyermark 6589* (CM); MCDONALD CO.: low ground, Noel, *Bush 5285* (US); STODDARD CO.: wooded slopes of ravines bordering east-facing escarpment on Crowley Ridge, just south of Stephens Cemetery, 1.5 miles s. of Pyletown, *Steyermark 66182* (CM); WAYNE CO.: wooded swamp in valley of Lost Creek, right along road, Shook P. O., *Steyermark 6572* (CM). **Arkansas.** ASHLEY CO.: Tehemanahought River bottoms, Hamburg, *Demaree 16379* (CM, NY); BRADLEY CO.: banks of Saline River, Warren, *Demaree 18383* (CM); FRANKLIN CO.: low moist areas, Lacey, *Demaree 23881* (NY); HEMPSTEAD CO.: low

woods, Tokio, *Demaree 9944* (NY); MONROE CO.: low bottoms, Brinkley, *Demaree 10885* (NY); NEVADA CO.: 4 miles s.e. of Prescott, *Hollister 121* (US); PIKE CO.: bottoms, Caddo River, Norman, *Demaree 9564* (G, NY). **Louisiana.** AVOYELLES PARISH: Marksville, *McAtee 2199* (US); BIENVILLE PARISH: sandy clay ditch in shallow water, 11 miles s.e. of Minden, *Shinners 12573* (SM); EAST BATON ROUGE PARISH: river banks, Oct. 1873, *Joor* (CM); NATCHITOCHES PARISH: low swampy woods, Natchitoches, *Palmer 8705* (NY); QUACHITA PARISH: edge of Bartholomew Bayou, *Bastrop 13840* (US); SAINT TAMMANY PARISH: Covington, *Bro. Arsène 11190* (US). **Oklahoma.** CREEK CO.: Sapulpa, *Bush 1446* (NY, US); JOHNSTON CO.: woods and fencerows, common, near Tishomingo, *Houghton 3522* (G); LEFLORE CO.: at edge of thicket, near pond, near Panama, *Stevens 2614* (G, NY, US); OKLAHOMA CO.: clay soil in open woods in overflow basin of the N. Canadian, 1 mile s. of Harrah, *Waterfall 3536* (G). **Texas.** BOWIE CO.: Texarkana, Oct. 1894, *Letterman* (NY, US); COLLIN CO.: 3 miles n.e. of Wylie, *Turner 1445* (SM); COMAL CO.: Comanche Spring, New Braunfels, *Lindheimer 953* (C, NY, SM, US); DALLAS CO.: in moist black clay of lake shore, Bachman's Lake near Lemmon Ave., *Whitehouse 22432* (SM); FREESTONE CO.: in pasture, low open woods, sandy soil, 12.5 miles e.s.e. of Fairfield on Highway 84, *Shinners 9638* (SM); MADISON CO.: in damp shady places, near Trinity River, *Dixon 428* (CM, G, NY); RED RIVER CO.: low grounds near creek, pine woodland, 9.5 miles n. of Clarksville on Highway 37, *Whitehouse 20525* (SM); VAN ZANDT CO.: near Ocean Lake n. of Edgewood, *Whitehouse 16449* (SM); WHARTON CO.: Pierce, *Tracy 7330* (CM, G, NY, US).

6a. *Pluchea purpurascens* (Sw.) DC, var. *purpurascens*, Prodr. 5: 452 (1836)

Conyza purpurascens Swartz, Prodr. 112 (1788). (As to descr. Browne, excl. syn. Sloane.)
C. odorata L., Syst. Nat. ed. 10, 1213 (1759). (As to plant described, not as to Sloane plate and single leaf of Plum. ed. Burm. plate on which name *C. odorata* was based.)
C. angustifolia Nutt., Jour. Acad. Nat. Sci. Phila. 7: 109 (1834); not *C. angustifolia* Roxb.
Pluchea glabrata DC., Prodr. 5: 452 (1836).
Stylimnus maritimus Raf. ex. DC. in syn., Prodr. 5: 452 (1836).
Pluchea camphorata sensu DC. (in part), Prodr. 5: 452 (1836).
P. camphorata sensu T. & G. var. *angustifolia* (Nutt.) T. & G., Fl. N. Am. 2: 261 (1838).
P. purpurascens (Sw.) DC., var. *glabrata* [incorrectly attributed to DC. by] Grisebach, Cat. Pl. Cub. 150 (1866).
Placus purpurascens (Sw.) Maza, Ann. Hist. Nat. Madrid 19: 273 (1890).
Pluchea camphorata sensu O. Kuntze var. *glabrescens* O. Kuntze, Rev. Gen. 1: 357 (1891).
P. camphorata sensu O. Kuntze var. *pubescens* O. Kuntze, *ibid.*
P. amorifera Badillo, Bol. Soc. Venezolana Cienc. Nat. 10: 309 (1946).

Annual herb, up to about 1.5 m. tall; stems glabrate below, copiously puberulent on the upper portions and on the inflorescence branches, the latter sometimes cinereous or sordid, leafy to the summit; leaves extremely variable: sessile or petiolate, generally short-ovate to ovate-lanceolate, sometimes lanceolate or elliptical, the apices generally obtuse varying in the more lanceolate or elliptical blades to long-acute, the margins from evenly to unevenly serrate or serrate-dentate to crenate-serrate or entire; surfaces of the blades varying from essentially glabrous or very sparsely puberulent to copiously puberulent or essentially tomentose; inflorescence cymose, the central axis maturing first, the laterals equalling or exceeding it in length, thus the inflorescence being flat-topped or layered; heads campanulate, 4.5–7 mm. high, 4.5–6 mm. broad; outer and median phyllaries copiously puberulent and ciliate, the inner sparsely puberulent on the summits, grading from ovate-obtuse on the outside of the involucres to oblong-obtuse in the median portions to linear-oblong and acute on the inside; the outer phyllaries herbaceous, the median and inner scarious and usually purplish or rose-colored; corollas rose-purplish. FIG. 16, PLATE 22.

Distribution: Salt or brackish marshes, salt flats or salt plains, and fresh, moist habitats, Maryland and Virginia to southern peninsular Florida and the Keys; westward through the Gulf States to Texas; in Arkansas, Kansas, and Oklahoma; from Texas to California, and in southwestern Utah and in Nevada; Mexico through Central America; in Columbia, Venezuela, and British Guiana; Bermuda, Bahama Islands, Greater Antilles, and the Leeward Islands.

Some specimens examined: **United States. Maryland.** CALVERT CO.: salt marsh, Chesapeake Beach, *Chase 2525* (CM); in marsh, Chesapeake beach, Aug. 26, 1904, *Tidestrom* (GA). **Virginia.** MIDDLESEX CO.: beach along estuary of Rappahannock River, Waterview, *Martin 1229* (NY); PRINCESS ANNE CO.: brackish to fresh marsh along Back Bay, at eastern margin of Long Island, *Fernald & Long 11190* (G, NY, US); YORK CO.: open marsh, Carters Creek, near York River, *Grimes 4268* (NY). **North Carolina.** BRUNSWICK CO.: Smith's Island, *Blomquist 5292* (D); BEAUFORT CO.: at edge of water, Pungo Creek, 6 miles n.w. of Belhaven, *Radford & Stewart 769* (NC); CURRITUCK CO.: sandy shore of Currituck Sound, at Point Harbor, *Godfrey & Fox 51014* (NCS); HYDE CO.: along wagon road at edge of salt marsh, edge of Pamlico Sound, 0.8 mile n. of Englehard, *Radford & Stewart 788* (NC); mud flat near shore, Lake Mattamuskeet, *Rudolph 8-77* (NCS). **South Carolina.** BEAUFORT CO.: St. Helena Island, border of salt marsh, Oct. 1903, *Cuthbert* (F); Beaufort District, 1883, *Mellichamp* (US); CHARLESTON CO.: Kiawah Island, Sept. 15, 1941, *Totten* (NC); shell plain, border of salt marsh, McClellanville, *Godfrey & Tryon 8147* (G, NY, US); GEORGETOWN CO.: Cat Island, *Alexander 127* (US). **Georgia.** Wilmington Island, *Hopkins 48* (NY); BRYAN CO.: Ossabaw Island, *Sperry 572* (US) CATHAM CO.: ditch near river, Savanna, *Eyles 3662* (GA); GLYNN CO.: Frederica Fort, S. Simons Island, *Ford & Thorne 2713* (NCS, T); low peaty pine barrens near salt marsh, 13 miles n.w. of Brunswick, *Wiegand & Manning 3284* (G); MCINTOSH CO.: Darien, *Biltmore Herbarium 4377e* (US); ditch near brackish marsh, near Eulonia, *Eyles 7592* (G); marshes of Altamaha River, 2 or 3 miles above Darien, *Harper 2000* (CM, G, NY, US). **Florida.** ALACHUA CO.: Newmans Lake, Gainesville, Sept. 10, 1940, *West & Arnold* (F); BREVARD CO.: in dry sandy soil along roadside, about 8 miles w. of Melbourne, *Moldenke 229a* (D, NY); CLAY CO.: hammock on river, West Tocoi, Sept. 6, 1937, *"Pasture Survey"* (F); COLLIER CO.: Collier-Seminole State Park, Feb. 18, 1946, *Beck* (F); DADE CO.: salt flat, near Chickey Key, Nov. 25, 1946, *Ledin* (F); DESOTO CO.; near Arcadia, *Schallert 12592* (F); DUVAL CO.: river shores near Jacksonville, *Curtiss 5107* (CM, NY, US); FRANKLIN CO.: St. Vincent Island, *Tracy 6361* (NC, NY, US); GADSDEN CO.: moist, rich ground, River Junction, *Curtiss 5990* (NC); GILCHRIST CO.: hammock along Santa Fe River, Oct. 1, 1943, *West & Arnold* (F); GULF CO.: swamps and low grounds, near Apalachicola, *Biltimore Herbarium 4377a* (G, NY, US) and *4377c* (G, NY, US); HAMILTON CO.: West Lake, in a swamp, July 16, 1929, *O'Neill* (F); HIGHLANDS CO.: moist pinelands, s. of Sebring, May 16, 1948, *Garrett* (F); HILLSBOROUGH CO.: moist, shaded ground, near the Bay, W. Tampa, March 28, 1933, *Churchill* (G); LAKE CO.: vicinity of Eustis, *Nash 1470* (C, CM, G, NY, US); LEE CO.: Sanibel Island, *Tracy 7247* (CM, NY, US); around ponds, Myers, *Hitchcock 160* (CM, G, NY, US); ORANGE CO.: shore, *Fredholm 5366* (G, US); PALM BEACH CO.: pinelands, Port Sewall, *Hunnewell 7427* (G); PINELLAS CO.: coastal ridge, Safety Harbor, *Blanton 6349* (G); Pinellas Point, St. Petersburg, Jan. 25, 1926, *Williams* (D); ST. LUCIE CO.: Eagle Bay, northern end of Lake Okeechobee, *Small & Small 4363* (NY); SUMTER CO.: wet ditch near Rutland Creek, 9 miles w. of Wildwood, Nov. 8, 1946, *West & Arnold* (F); VOLUSIA CO.: bank of Tomoka River, entered by Tomoka Ave., Ormond, Aug. 22, 1946, *Butts & Arnold* (F).

Mississippi. Bird Island, the Mississippi Delta and Adjacent Islands, *Lloyd & Tracy 497* (NY); HARRISON CO.: Biloxi, *Tracy 4754* (C, CM, G, NY, SM, US); JACKSON CO.: Ocean Springs, *Seymour 91929* (D). **Arkansas.** HEMPSTEAD CO.: common along river, Fulton, *Bush 1017* (NY, US); PULASKI CO.: sand bars, Arkansas River, Woodson, *Demaree 8624* (G, US). **Louisiana.** CALCASIEU PARISH: vicinity of Lake Charles, *Allison 107* (NY, US); IBERIA

PARISH: in salt marsh, Avery Island, *Correll & Correll 9538* (D, G); NATCHITOCHES PARISH: along edge of shaded pool on bluff above river, Red River, Grand Encore, *Ewan 17516* (GA); WEBSTER PARISH: in wet sandy soil, edge of stream, 4 miles w. of Minden, *Correll & Correll 10511* (D, G). **Kansas,** SEDGWICK CO.: moist places, *Clothier 1092* (G, NY, US). **Oklahoma.** ALFALFA CO.: in deep sand, north edge of Salt Plains Lake, east of Cherokee, *Waterfall* 9771 (SM); CLEVELAND CO.: river flats, 6 miles n.w. of Norman, Oct. 4, 1924, *Bruner* (US); LOGAN CO.: sandy banks of Cimaroon River, near Langston, *Olive 63* (CM). **Texas.** BELL CO.: near Belton, *Wolff 1313* (US); BEXAR CO.: Blue Lake, 14 miles s.e. of San Antonio, *Schulz 389* (G, US); moist soil near San Antonio, *Biltmore Herbarium 4377j* (US); Helotes Creek bank, *Sister Mary Clare, C. D. P. 368* (NY); BLANCO CO.: springy river banks, Blanco, *Palmer* 12870 (NY); CAMERON CO.: Brownsville, *Runyon 444* (G) and *2007* (CM); COMANCHE CO.: infrequent in moist soil along borrow ditch, bottoms of Sabina Creek, 3⅓ miles s. of De Leon, *Cory 58145* (SM); COOKE CO.: edge of Sarles Lake, 3 miles s. of Gainesville, *McCart 1835* (NY); DONLEY CO.: margin of Lelia Lake on Highway 287, *Whitehouse 17259* (SM, US); EL PASO CO.: *Cory 2516* (G); GALVESTON CO.: in marshy soil; prairies, about 1 mile w. of Galveston, *Whitehouse 10973* (SM); GILLESPIE CO.: Palo Alto, *Jermy 802* (US); Fredericksburg, *Dapprich 6182* (SM); GONZALES CO.: marsh, *Tharp 127* (G); HAYS CO.: San Marcos and vicinity, summer 1897, *Stanfield* (NY); HENDERSON CO.: frequent in woods along small drainage course, 9⅔ miles n.w. of Athens, *Cory 49726* (SM); HOPKINS CO.: sandy stream bottom pasture, 7.8 miles s. of Sulphur Springs, *Shinners 11681* (SM); JOHNSON CO.: infrequent in woods at west edge of lake, Cleburne State Park, 1.5 miles n.w. of entrance, *Cory 58016* (SM); KENDALL CO.: wet open ground, Boerne, *Palmer 12276* (C, NY, US); KENEDY CO.: El Toro Island, mud flats of the Laguna Madre, *Tharp 49099* (C, US); MCLENNAN CO.: in marshy limestone soil, about 2 miles s.w. of Waco on old Temple Road, *Whitehouse 10978* (SM); PECOS CO.: along the border of the reservoir at Leon Springs about 10 miles w. of Ft. Stockton, Oct. 3, 1948, *Hinckley* (US); SAN PATRICIO CO.: ½ mile north of Aransas Pass, in sandy type of soil near roadside ditch, *Jones 540* (SM); TARRANT CO.: open woods, Handley, *Ruth 246* (NY); UVALDE CO.: Nueces River, 4 miles s. of Camp Wood, *Cory 38221* (G); VAN ZANDT CO.: 3 miles e. of Grand Saline on Highway 80, *Whitehouse 16469* (SM); WILLIAMSON CO.: along margin of creek about 6.5 miles w. of Round Rock, *York 46321* (D, NC); WOOD CO.: low swampy grounds near creek, 4.5 miles e. of Winnsboro, *Whitehouse 17591* (SM). **New Mexico.** CHAVES CO.: Roswell, alt. 3800 ft., *Earle & Earle 328* (C, G, NY, US); DONA ANA CO.: valley of the Rio Grande, below Dona Ana, *Parry et al. 532a* (NY, US); EDDY CO.: along ditch through gravelly ridge, 8 miles n. of Artesia, *Waterfall 4274* (G); south of Carlsbad, *Standley 40350* (US). **Arizona.** Akaline flat along the flood wash of the Colorado in the vicinity of Yuma, *Sister Mary Noel Fochtman, O. S. F. 34* (NY); low wash near roadside on the way to Welton, e. of the Gila Mountains, *Sister Mary Noel Fochtman, O. S. F. 49* (NY); Bill Williams Mt., 1869, *Palmer* (US); near mouth Agua Fria, *Peebles et al. 220* (US). **Utah.** WASHINGTON CO.: low ground, Washington, *Hall 562* (US). **Nevada.** Watkins Ranch, Ash Meadow, alt. 720 m., *Coville & Funston 346* (US); Telescope Mt., *Wheeler* (G). **California.** Dos Palms, Colorado Desert, *Schellenger 23* (C); Palmas, Colorado Desert, Jan. 21, 1921, *Childs* (C) SANTA CATALINA ISLAND: Avalon, Aug. 1897, *Trask* (US); in a wet spot in Soapstone Quarry, *Nuttall 904* (CM); S. BERNANDINO VALLEY: damp saline soil, *Parish Bros. 567* (G, US); ALAMEDA CO.: Strawberry Canyon in Berkeley Hills, open ground along the trail, Mar. 28, 1928, *Clausen* (DN); CONTRA COSTA CO.: in tule swamp, Martinez, region of San Francisco Bay, *Hall 10272* (C); LOS ANGELES CO.: Cienega, near Los Angeles, damp ground, *Blake 862* (CM, G); ORANGE CO.: *Zumbro 381* (C); Bolsas Marsh, *Booth 1272* (C); RIVERSIDE CO.: Salton, *Davy 8038* (C); SAN BERNARDINO CO.: swampy ground near Rincon, *Munz 10808* (C); SAN DIEGO CO.: Del Mar, Aug. 6, 1906, *Brandegee* (C); SAN JOAQUIN CO.: Holt, *Mason 5419* (C); SOLANO CO.: salt marshes, Suisun, *Bolander 2622* (C, G, US); TULARE CO.: between Tulare and Tulare Lake, *Palmer 2765* (US); VENTURA CO.: Oxnard, *Davy 7816* (C).

Mexico. TAMAULIPAS: vicinity of Tampico, alt. about 15 m., *Palmer 138* (CM, G, NY, US); VERA CRUZ: Antigua, *Purpus 6034* (C, CM, G, NY, US); COAHUILA & NUEVO LEÓN:

Mts. of Montclara, *Palmer 544* (CM, G, US). BAJA CALIFORNIA: Todos Santos Bay, *Orcutt 1236* (CM, G, US). NAYARIT: Mexcaltitan, *Ortega 5538* (US); San Blas, Tepic, *Maltby* 18 (US). OAXACA: Tehuantepec City, *Nelson 2612* (G, US). CAMPECHE: in swamp-side, Palizada, *Matuda 3859* (CM, G, NY). YUCATÁN: Izamal, *Gaumer 1002* (G, NY); Chichankanab, *Gaumer 2175* (CM). COZUMEL ISLAND: Caleta, *Millspaugh 1532* (CM). CHIAPAS: Paderón, Tonala, *Matuda 16278* (US).

British Honduras: Prospecto, Northern River, *Gentle 860* (C, CM, NY, US). **Honduras:** in bog near seashore, vicinity of Ceiba, *Yuncker et al. 8232* (G, NY). **Nicaragua:** Managua, *Chaves 54* (US). **Panama:** Barro Colorado Island, Canal Zone, on floating island, large cove e. of Redwood House, *Woodworth & Vestal 611* (CM, G).

Colombia: Puerto Colombia, *Bro. Paul 856* (CM); Santa Marta, *Smith 1995* (CM, G, NY, US). **Venezuela:** Catia del Mar, Lugar, planta de la playa, *Padre Vogl 627* (CM). SUCRE: hot springs near La Toma, between San Juan and Cumaná, southeast of Cumaná, alt. 60 m., *Steyermark 62860* (Type collection of *Pluchea amorifera* Badillo: CM (type), G, NY (isotypes). ISLAND OF MARGARITA: El Valle, *Miller & Johnston 1* (CM). **British Guiana.** *Jenman 2190* (NY).

Bermuda: shade of *Rhizophora Mangle* L. and *Avicennia nitida* Jacq., Pembroke Marshes, Pembroke Parish, *Moore 3162* (G). **Bahama Islands.** GREAT BAHAMA: clearing, Eight Mile Rocks, *Britton & Millspaugh 2598* (CM, NY); BIMINI ISLAND GROUP: South Bimini, *Howard & Howard 10026* (SM, US); CAT ISLAND: roadside, the Bight and vicinity, *Britton & Millspaugh 5927* (CM, NY); GREAT EXUMA: sink holes near Georgetown, *Britton & Millspaugh 3102* (CM, NY). **Cuba.** ISLE OF PINES: near Nueva Gerona, *Palmer & Riley 968* (US), *866* (NY). PINAR DEL RÍO: marsh, Corrientes Bay, *Britton & Cowell 9930* (NY); *Havana*: banks of a rivulet, near Guanabacoa, *Bro. León 2546* (NY). **Matanzas:** Cárdenas, *Britton & Wilson 199* (NY). Las Villas: plain, Río San Juan, *Britton et al. 5948* (NY). ORIENTE: banks of Río de Ymias (?), southern Baracoa Region, *Fre. León 12163* (NY). **Jamaica:** marsh, Flat Point, Saint Ann's Bay and vicinity, *Britton 2526* (NY). **Haiti:** Bayeux, near Port Margot, *Nash 323* (NY); vicinity of Étang, Étang Saumatre, *Leonard 3540* (NY, US). **Dominican Republic.** MONTE CHRISTI: coastal barrens and elevated coral reef near Puerto Libertador, Manzanilla Bay, *Howard & Howard 9641* (G). SANTIAGO: Mao, *Abbott 1066* (US); BARAHONA: Cabral, *Abbott 2000* (G, US); AZUA: Azua, *Rose et al. 4007* (US). SANTO DOMINGO: wet soil of Ozama River flats, 2–3 miles from mouth, vicinity of Ciudad Trujillo, *Allard 15905* (G). **Puerto Rico:** Río Piedras, *Johnston 580* (NY); coastal marsh, Humacao, *Sargent 3005* (US); Mona Island, *Stevens 6165* (NY); dry serpentine slope, Guanajebo, *Britton & Britton 9392* (NY); marshes, Laguna Tortuguero, *Britton & Britton 9241* (CM, NY); pastures, Dominguito, *Cowles 207* (US); Pueblo Beajo, Catano, *Goll 853* (US); Guanica, Lake, *Sargent 7* (US); TÓRTOLA: swamp, Zion Hill, *Fishlock 438* (NY, US); ANEGADA: rocky plain near settlement, *Britton & Fishlock 1012* (NY, US). **St. Croix:** Blackwood's Estate, *Ricksecker 376* (C, CM, G, NY, US). ANGUILLA: *Boldingh 3492B* (NY). ST. MARTIN: *Boldingh 2621B* (NY). **Guadeloupe:** Marais du Gosier, pes Loussette, *Quentin 305-W* (G); *Questel 2464* (US); *Père Duss 2953* (DN, NY); *Stehle 305* (US).

6b. *Pluchea purpurascens* (Sw.) DC., var. *succulenta* Fern., Rhodora **44**: 227 (1942)

Conyza marilandica Michx., (?), Fl. Bor.-Am. **2**: 126 (1803).
Pluchea marilandica (Michx.) Cass., (?), Dict. Sci. Nat. **42**: 2 (1826).
P. marilandica sensu DC. (in part), Prodr. **5**: 451 (1836).
P. camphorata sensu DC. (in part), Prodr. **5**: 452 (1836).

Annual herb, up to about 6 dm. tall; stems stout, glabrate below, copiously puberulent on the upper portions and on the inflorescence branches, leafy to the summit; leaves sessile or with very short petioles, elliptical to rhombic ovate-anceolate, occasionally obovate or oblanceolate, obtuse or rarely acute, margins

serrate-dentate to entire; median leaves from *ca.* 3 cm. long and 1.5 cm. broad on some plants to *ca.* 10 cm. long and 4 cm. broad on others; leaves succulent (drying thin), the surfaces from glabrous to copiously puberulent above and below; inflorescence with few to many heads in panicled cymes, more or less flat-topped; heads broadly campanulate, *ca.* 8–10 mm. high, 7–10 mm. broad; outer and median phyllaries copiously puberulent and ciliate, the inner sparsely puberulent on the tips and somewhat ciliate, grading from oblong-acuminate on the outside of the involucres to oblong or oblong-elliptical in the median portions and linear-oblong on the interior; phyllaries often suffused with rose-purple color; corollas rose-purplish. FIG. 9, PLATE 22.

Distribution: salt and brackish marshes along the coast from eastern North Carolina to northeastern Massachusetts, salt areas in southeastern Michigan, and in southern Illinois.

Type Specimen: ditch in old marsh, Back Bay, Boston, Massachusetts, Sept. 17, 1910, *E. F. Williams* (G).

Some additional specimens examined: **North Carolina.** CARTERET CO.: salt marsh, Cape Lookout, near lighthouse, *Quarterman 346* (D). **Virginia.** ELIZABETH CITY CO.: brackish marsh, w. of Hampton, *Fernald et al. 5114* (G); NEW KENT CO.: salt marsh by York River, back of Terrapin Point, n.e. of Holly Forks, *Fernald & Long 11633* (G); MIDDLESEX CO.: beach along estuary of Rappahannock River, Waterview, *Martin 1229* (D); NORFOLK CO.: Norfolk, *Heller 740* (C, CM, G, NY); NORTHAMPTON CO.: Cape Charles City, *Canby & Rose 846* (US); Smith's Island, Oct. 1, 1897, *Palmer* (US); PRINCESS ANNE CO.: fresh to brackish swales along North Landing River, near Creed's, *Fernald et al. 5113* (G, NY); YORK CO.: in tidal marsh of creek, Capitol Landing, Queen's Creek, n. of Williamsburg, *Grimes 4292* (G); DISTRICT OF COLUMBIA: vicinity of Washington, *McCarthy* (CM). **Maryland.** ANNE ARUNDEL CO.: colonies along water's edge, South River near confluence of Beard Creek, *Ewan 17353* (GA, SM); CALVERT CO.: sandy marsh behind Chesapeake Beach, *House 351* (NY); DORCHESTER CO.: salt marsh, near Cambridge, *House 243* (NY); ST. MARY'S CO.: Tall Timbers, *Killip 32199* (US); TALBOT CO.: marsh on east shore of Poplar Island, Chesapeake Bay, *Allard 7429* (G, US); WORCHESTER CO.: brackish marsh, 1.75 miles n. of Ocean City, *Fogg 11410* (G). **Delaware.** KENT CO.: along margin of Little Creek, e. of Little Creek, *Larsen 360* (G); Kitts Hummock, *Larsen 346* (D); marsh near Beach, ½ mile s. of Smyrna River, *Larsen 1043* (GA); NEW CASTLE CO.: in brackish swamp along Delaware Bay, Port Penn, Sept. 10, 1927, *Baker* (D); SUSSEX CO.: border of brackish marsh, by Indian River, Oak Orchard, *Fernald & Long 4250* (G). **Pennsylvania.** LANCASTER CO.: on Piney Island, Susquehanna River, Sept. 23, 1913, *Otis* (NY); PHILADELPHIA CO.: commons, below the Navy Yard, Philadelphia, Sept. 25, 1864, *Diffenbaugh & Porter* (G); low ground, below the Navy Yard, Philadelphia, Sept. 28, 1864, *Parker* (NY). **New Jersey.** ATLANTIC CO.: salt marshes, Atlantic City, Sept. 29, 1885, *Brinton* (CM); BERGEN CO.: salt marsh, Newark, Aug. 25, 1896, *Lyon* (US); marshes near Newark, *Rusby* (F); HUDSON CO.: Weehawken, Sept. 14, 1895, *Van Sickle* (US); Hoboken, Oct. 2, 1847, *Holton* (CM); SALEM CO.: border of brackish marsh about 4 miles s. of Canton, *Fogg 6047* (G). **New York.** QUEENS CO.: Rockaway Beach, Sept. 1892, *Jelliffe* (CM); RICHMOND CO.: in salt marsh, Arthur Kills, Staten Island, *Gershoy 851* (G); ROCKLAND CO.: patches in *Spartina* marsh, Piermont, *Smith & Birdsey 3442* (GA); SUFFOLK CO.: Montauk Point, L. I., *Barnhart 1156* (NY); WESTCHESTER CO.: brackish marsh, north Tarrytown, *Barnhart 1655* (NY). **Connecticut.** FAIRFIELD CO.: common in salt meadows, Stratford, Aug. 16, 1893, *Eames* (C, US); salt marshes, Bridgeport, Aug. 27, 1891, *Johnson* (D); salt marsh, Stratford, Sept. 16, 1905, *Eames* (NE); NEW HAVEN CO.: Madison, Aug. 17, 1874, *Hall* (NE); NEW LONDON CO.: Norwich, Aug. 26, 1885, *Setchell* (NY). **Rhode Island.** KENT CO.: marsh, Sandy Point, Warick, Sept. 17, 1922, *Collins* (NE); NEWPORT CO.: salt marshes about Great Salt Pond, Block Island, *Fernald et al. 10627* (G, NY); Tiverton, Aug. 5, 1877, *Phillips* (NE); PROVIDENCE CO.: near Eden Park, Cranston, Sept. 9, 1909, *Hope* (NE).

Massachusetts. BARNSTABLE CO.: brackish marsh, West Barnstable, Sept. 17, 1916, *St. John & Hunnewell* (NE); BRISTOL CO.: salt marsh, So. Somerset, Aug. 8, 1913, *Sanford* (NE); DUKES CO.: salt marsh, Edgartown, Martha's Vineyard, Sept. 11, 1901, *Fernald* (*PLANTAE EXSICCATAE GRAYANAE 70*) (C, CM, DN G, GA, NE, NY); ESSEX CO.: edge of salt marsh Ipswich, near Daland's Camp, Sept. 2, 1921, *Morse* (NE); MIDDLESEX CO.: marsh back of Stillman Infirmary, Cambridge, Aug. 27, 1912, *Deane* (NE); NORFOLK CO.: salt marsh, Cohasett, Aug. 19, 1882, *Deane* (NE); salt marsh, Scituate, Sept. 13, 1914, *Knowlton* (NE). **Michigan.** salt grounds at River Rouge, *Farwell 8758* (G); WAYNE CO.: near salt wells and mine at Oakwood, West Detroit, Sept. 12, 1914, *Chandler* (US). **Illinois.** UNION CO.: Jonesboro, *Hall* (CM).

Pluchea purpurascens (*sensu lat.*) is a very wide ranging plant. In this study quite a large number of specimens was examined, representing the population from throughout its range. Habitat data on the specimen labels is meagre, as is the author's knowledge of the ecological and geographical factor differences operating over so wide an area. Within these limitations, it is possible to make only a very general statement relative to the habitat relations of the populations in different parts of the range. In the eastern United States, from Georgia northward, this plant apparently is confined to brackish or saline habitats. In other parts of the range it may occur in saline to fresh, moist habitats, or in saline or alkaline areas in the more arid regions of the West. Its extensive geographical range should be emphasized. It occurs from the north temperate eastern United States southward through sub-tropical and wet tropical areas of the southeastern United States and much of the West Indies, westward to the more arid regions of the southwestern United States and Mexico, southward through wet tropical Mexico and Central America and the northern coastal regions of South America.

In general, this species exhibits a considerable degree of variation in foliar characters, in pubescence, and in size of heads. An analysis of the population, confined as it was to herbarium material, revealed but one set of morphological variations, apart from the great mass of material, which could be reasonably well correlated with geography, namely the portion of the population distinguished as *P. purpurascens*, var. *succulenta*. It will be recalled from the earlier discussion in this paper that this was Fernald's conclusion when he named the variety *succulenta*. He recognized then, for the first time, that plants, which in various parts of the range were being called both *P. purpurascens* and *P. camphorata*, were indeed members of one species-population.

Relative to the portion of this population here treated as var. *purpurascens*, or the typical, plants occurring in the more arid regions of the southwestern United States and northern Mexico possibly could be varietally segregated. They have somewhat larger heads than plants from other areas, though they are not nearly as distinctive in this regard as var. *succulenta*, and are more copiously and uniformly pubescent. These, however, are differences of degree and they "fuzz-off" so gradually and imperceptibly, both to the eastward and to the southward, that at present no definite conclusions are drawn. In this regard, it is suggested that this whole variable population, as widely distributed as it is geographically, and apparently also ecologically, might represent an excellent source of material for a detailed experimental analysis.

It should be emphasized, too, that, although *P. purpurascens*, var. *succulenta*

is clearly recognizable as a morphological entity to the north of Maryland, from Maryland southward to North Carolina it is often difficult to distinguish specimens as being the more nearly like var. *succulenta* or var. *purpurascens*. In this region the characters intergrade. However, plants from the isolated stations in southeastern Michigan and southern Illinois are quite characteristic var. *succulenta*. Incidentally, Fernald, in Gray's Manual ed. 8, (1950), does not record the occurrence of var. *succulenta* in these stations in states widely removed from the northeastern coastal areas.

Pluchea amorifera Badillo, described in 1946 from Venezuela, appears to be not unlike *P. purpurascens*, var. *purpurascens* in character except that the specimens on which the name is based are depauperate.

7. *Pluchea longifolia* Nash, Bull. Torr. Bot. Club **23**: 108 (1896)

Perennial herb, 6–12 dm. tall; stems commonly dark-purplish, glabrate below, sparingly pubescent in the median portions, grading to lanate on the ultimate inflorescence branches; leaves large, overlapping, sessile, the lower narrowed to truncate-cuneate bases, gradually becoming cordate-clasping above, varying from broadly oblong-lanceolate below, through oblong or oblong-elliptical to ovate-oblong, the apices generally obtuse, the margins irregularly and coarsely serrate, the teeth apiculate, scabrous above, sparsely short-pubescent underneath, or the uppermost copiously short-pilose below, especially on the veins; inflorescence of few to numerous heads in relatively loose cymes; heads conspicuously turbinate, *ca.* 10–12 mm. high and ca. 7–8 mm. broad in the median portions of the involucres; phyllaries green, herbaceous, short-pubescent, more or less short-ciliate or short-fimbriate; the lowermost phyllaries very short, ovate, obtuse-apiculate and actually beginning somewhat down on the peduncles, becoming longer and broader through oblong, broadly obtuse-apiculate, the inner linear-oblong or linear-attenuate; corollas creamy white. FIG. 7, 8, PLATE 21.

Distribution: Swamps, marshes, and lake shores from south-central peninsular Florida to northeastern Florida.

Type collection: BREVARD CO.: Titusville, July 30 to Aug. 1, 1895, *Nash 2293* (CM, G NY, US). Although not so designated by Nash, the type specimen is here taken to be the one deposited at the New York Botanical Garden Herbarium.

Some additional specimens examined: DUVAL CO.: near Mayport, *Lighthipe 332* (NY); FLAGLER CO.: high hammock, 6 miles e. of Bunnell, Oct. 10, 1940, *West & Arnold* (F); HERNANDO CO.: June-July 1898, *Hitchcock* (CM); LEVY CO.: ditch bank, Gulf Hammock, July 21, 1937, *West & Arnold* (D, F); open places around pond, Gulf Hammocks, *Murrill 636* (US); MANATEE CO.: *Simpson* (US); PUTNAM CO.: wooded swamps of Mud Springs, Welaka, Oct. 27, 1940, *Laessle* (F); ST. JOHN CO.: St. Augustine, *Biltmore Herbarium 4185b* (US); SUMTER CO.: marsh at Withlacoochee River bridge near Rutland, Nov. 8, 1946, *West & Arnold* (F); VOLUSIA CO.: edge of swamp, Oak Hill, *Curtiss 6700* (C, G, NY, P, SM, US); open pinelands near Oak Hill, *Small et al 11549* (F, G, NY, US); "N. FLORIDA": swamp of Wakulla River at Lower Bridge, *Small, Harper & Gunter 9679* (D, NY, T).

8a. *Pluchea foetida* (L.) DC., var. *foetida*, Prodr. **5**: 452 (1836)

Baccharis foetida L., Sp. Pl. ed. **1**: 861 (1753). (As to plant of Gronovius; excluding synonym and figure of Dillenius.)
B. viscosa Walt., Fl. Carol. 202 (1788), in part, not Persoon.

Conyza amplexicaulis Michx., Fl. Bor.-Am. **2**: 126 (1803); not *C. amplexicaulis* Willd.
C. uliginosa Pers., Syn. Pl. **2**: 427 (1807). (Renaming of *C. amplexicaulis* Michx., not Willd.)
Pluchea bifrons DC. Prodr. **5**: 451 (1836); not *Conyza bifrons* L.
P. foetida (L.) B.S.P., Prelim. Cat. Fl. N. Y. (1888).
Placus bifrons (DC.) Gomez, Ann. Hist. Nat. Madrid **19**: 273 (1890).
Pluchea Eggersii Urban, Symb. Ant. **3**: 407 (1903).
P. tenuifolia Small, Man. Southeast. Fl. 1399 (1933).

Perennial herb, 5–8 dm. high; roots thick-fibrous; stems spongy-thickened at base, commonly dark-purplish, glabrate below, sparingly pubescent in the median portions, densely pubescent and approximately lanate on the inflorescence branches; leaves 3–10 (rarely up to 13) cm. long, 1–3 cm. broad, sessile; the lower cauline leaves lanceolate or oblanceolate with cuneate or truncate bases; median and upper leaves oblong, oblong-elliptical, occasionally ovate-oblong, rarely lanceolate-elliptical, typically shallowly auriculate-clasping, but occasionally the median and upper leaves as well as the lower with cuneate bases; leaf margins shallowly and unevenly apiculate-serrate; leaf surfaces glandular above, pubescent below; inflorescence of numerous heads in loose to (rarely) congested panicled cymes which tend to be flat-topped; heads broadly campanulate, *ca.* 8 mm. high; phyllaries green, herbaceous, pubescent and with ciliate margins, loosely imbricate, the outermost lanceolate-ovate with obtuse to obtuse-apiculate apices, the median oblong with acute apices, the inner linear, acute to short-attenuate; corollas creamy-white. FIGS. 1 & 2, PLATE 20.

Distribution: Wooded or open, wet areas: swamp forests, flatwoods, shrub-bogs, swales, ditches, borrow pits, and pond margins; chiefly on the coastal plain, from southern New Jersey to the Lake Okeechobee region of peninsular Florida, to eastern Texas, in southern Missouri (?), and in the Dominican Republic, West Indies.

Type Specimen: Clayton No. 159 from Virginia (in the Herbarium of the British Museum of Natural History). A photograph of the Clayton specimen was very kindly supplied by Mr. G. Taylor, Keeper of Botany, British Museum.

Some additional specimens examined: **United States. New Jersey.** CAPE MAY CO.: marsh, Cape May, Aug. 23, 1877, *Parker* (G, NY). **Delaware.** Cypress swamp, Gumboro, Aug. 15, 1877, *Commons* (P); SUSSEX CO.: moist hollows in sand dunes, s. of Bethany Beach, *Fogg 11257* (CM). **Maryland.** SOMERSET CO.: sandy pineland, 2–3 miles n. of Princess Anne, *Pennell 16105* (P); WICOMICO CO.: meadow above Salisbury, *Wherry & Pennell 12841* (NY, P); Salisbury, Sept. 1863, *Canby* (C, CM). **Virginia.** NORFOLK CO.: near Ocean View, *Kearney 2394* (US); NORTHAMPTON CO.: Cape Charles City, *Canby & Rose 813* (US); moist, peaty depression in pine woods s. of Townsend, *Fernald et al. 5545* (G); SOUTHHAMPTON CO.: wet siliceous and argillaceous drained border of Predler's Pond, Nottoway Swamp, s.w. of Sedley, *Fernald & Long 7688* (G). **North Carolina.** BEAUFORT CO.: marsh near Washington, *Godfrey 5393* (G); BERTIE CO.: roadside ditch, 6 miles n.e. of Windsor, *Fox 4093* (D, F, G, NA, NCS, NY); BLADEN CO.: ditch, White Lake, *Godfrey 5965* (D, G); BRUNSWICK CO.: abundant in shrub-bog, near Maco, *Godfrey 50419* (CM, D, DN, K, NA, NCS, NY, SMU); CRAVEN CO.: New Bern, *Kearney 1972* (US); CUMBERLAND CO.: in dry field near Rock Fish Creek, about miles s.e. of Fayetteville, Oct. 8, 1939, *Radford & Stewart* (NC); CURRITUCK CO.: marsh, Sligo, *Godfrey 5285* (G, US); DARE CO.: peaty depressions in sand flats, vicinity of Kill Devil Hill, *Godfrey & Fox 51032* (CM, D, DN, NA, NCS, NY, T); PITT CO.: wet roadside ditch, 1.5 miles s.w. of Stokes, *Godfrey et al. 50672* (NA, NCS, NY); ROBESON CO.: dried floor of *Taxodium-Nyssa* swamp, Lumber River, 3.5 miles s.e. of Pembroke, *Fox 5552* (NCS); WAKE CO.: between marsh and upland, Parten's Mill, *Godfrey 2334* (D, G, NCS). **South Carolina.**

BEAUFORT CO.: swale, Garden's Corners, *Godfrey 50940* (CM, D, NA, NCS, SM); BERKELEY CO.: swamp forest, south of Jamestown, *Godfrey & Boyce 50471* (CM, D, DN, F. K, NA, NCS, T); GEORGETOWN CO.: abundant in a small pool in pine flatwoods, 4.5 miles w. of Georgetown, *Godfrey & Boyce 50433* (CM, D, DN, F, GA, NA, NCS, NY, SM, T); MARLBORO CO.: in small swampy woodland, 8.7 miles n. of Cheraw, *Godfrey & Fox 50573* (D, F, NA, NCS); ORANGEBURG CO.: Eutawville, *Eggleston 5026* (NY). **Georgia.** BERRIEN CO.: sandy soil, open pine woods, 8 miles n.n.e. of Nashville, *Duncan 11823* (GA); CHARLESTON CO.: St. Mary's River swamp, below Trader's Hill, July 24-26, 1895, *Small* (CM, NY); EVANS CO.: open, mixed woods, wet, 5 miles e. of Clayton, *Duncan 2748* (F, GA, T); JENKINS CO.: swampy place in pineland, near Buckhead Church, 2.5 miles n. of Millen, *Pyron & McVaugh 915* (GA); LOWNDES CO.: open pine woods, 13 miles n. of Valdosta, *Quarterman 124* (D); WORTH CO.: drainway of open pinelands, 10 miles s.w. of Sylvester, *Duncan 2949* (GA).

Florida. ALACHUA CO.: swamp in shade along Sugarfoot Prairie, Gainesville, Oct. 31, 1937, *Watson* (F); BREVARD CO.: swamp, Okeechobee Region, *Fredholm 5980* (G, NY, US); CALHOUN CO.: river bottom, Altha, Sept. 9, 1931, *West* (F); CLAY CO.: in dry sandy soil, about 2 miles n. of Orange Park, *Moldenke 5241* (NY); DESOTO CO.: flatwoods, Lacy, Sept. 23, 1938, *West* (F); DIXIE CO.: Oldtown, *Howell 1126* (CM); DUVAL CO.: moist pine barrens, near Jacksonville, *Curtis 1374* (C, CM, G, NY, US); HARDEE CO.: 5.5 miles n. of Bowling Green on road to Fort Mead, *Hand 360* (NY)—the Type of *Pluchea tenuifolia* Small; HERNANDO CO.: Brooksville, June 27, 1934, *Reed* (F, NA); HILLSBOROUGH CO.: 27 miles s. of Tampa, June 24, 1934, *Reed* (NA); LEVY CO.: hammock, s.e. of Rosewood, July 10, 1937, *"Pasture Survey"* (F); MARION CO.: riverbank, Orange Springs Landing, Sept. 16, 1941, *West & Arnold* (F); OKALOOSA CO.: from Turkey Creek Swamp, *Tyson 268* (F); WASHINGTON CO.: damp mixed woods, near Vernon, *Hood 2822* (F). **Alabama.** BALDWIN CO.: along road, Mobile Delta, *Lueth & Blomquist* 128 (D); pine barrens, 5 miles n. of Bay Minette, Aug. 17, 1937, *LeClair* (NC); in low pineland, Perdido, *Blanton 68* (CM, G, NA, NY, US); BUTLER CO.: wet soil near Greenville, *Biltmore Herbarium 2956d* (US); ELMORE CO.: Deatsville, *Pollard & Maxon 303* (G, GA, US); LEE CO.: Macon's Mill, *Earle 853* (NY); MOBILE CO.: ballast, Hunter's Wharf, Mobile, 1899, *Mohr* (NY); TALLAPOOSA CO.: Thornton, *Earle 3130* (G, NY). **Mississippi.** Along ravines, Port Christian, *Langlois 268* (C); HARRISON CO.: vacant lot, Mississippi City, *Hood 338* (F); Biloxi, *Lloyd & Tracy 548* (NY); PEARL RIVER CO.: McNeill, *Reed 455* (NA); JACKSON CO.: wet swampy areas, Ocean Springs, *Demaree 28091* (SM). **Missouri.** BUTLER CO.: Poplar Bluffs, Aug. 5, 1879, *Letterman* (NY, P, US). **Arkansas.** DREW CO.: swamp, in shade, Wilmar, *Demaree 24599* (G, NY). **Louisiana.** CALCASIEU PARISH: low woods, Lake Charles, *Palmer 8520* (NY); RAPIDES PARISH: Alexandria, *Hale* (G, NY, P); moist soil on edge of small seepage pool, n. edge of Pineville, *Correll & Correll 9897* (D, G, NA); SAINT TAMMANY PARISH: pine woods, flowers whitish-yellow, Madisonville, *Riddell* (P); vicinity of Covington, *Bro. Arsène 11014* (US); TANGIPAHOA PARISH: moist, open grassy soil, 3 miles e. of Robert, *Correll & Correll 9315* (D, G, NA). **Texas.** ARANSAS CO.: Aransas Refuge, 0.5 mile w. of Alamos Well, *Uzzell 168* (NA); AUSTIN CO.: 7 miles n.e. of Bellville, *Tharp 42-125* (G); HARDIN CO.: 5 miles s. of Silsbee, *Cory 20050* (NA); HARRIS CO.: Harrisburg Sept. 1775, *Joor* (P); Hockley, 1890, *Thurrow* (CM); HENDERSON CO.: in mud, in damp thicket, with *Baccharis*, *Myrica*, and *Mikania*, 5.5 miles s.s.e. of Athens, *Shinners 9607* (SM); ROBERTSON CO.: bog, *Barkley 284* (NY); SMITH CO.: swamps, Swan, *Reverchon 3306* (SM); UPSHUR CO.: frequent in woods at edge of swamp along creek, 4.75 s.e. of Gilmer, *Cory 57709* (SM).

Dominican Republic. Savanna de Cupey, 360 m., June 30, 1887, *Eggers 2681* (K)—the Type collection of *Pluchea Eggersii* Urban; Sasua coastal swamp at Cabarete Cordillera Septentrional, Puerto Plata, *Ekman 14543* (US).

Although the identity of *Baccharis foetida* L., upon which *Pluchea foetida* (L.) DC. rests, has been established, in the general discussion of this species in relation to others of the complex, certain other points require clarification.

The Linnean treatment included two dissimilar elements: (1) the Gronovian-

Clayton, which, since it is quoted first in a positive fashion and is based upon an actual Clayton specimen, is taken as the type basis; and (2) a Dillenian reference[21] which Linnaeus himself questioned. The Dillenian plate is clearly unlike the Clayton plant. It is identifiable with *Erigeron camphoratum* L. (*Pluchea camphorata* (L.) DC.).

When de Candolle transferred *Baccharis foetida* to *Pluchea*, he identified the name with the Dillenian plate, which Linnaeus had questioned, and entirely ignored the Gronovian-Clayton element. Thus, in making the transfer he misidentified the plant, but by present rules the combination stands. It belongs to the same plant to which the name was originally given; it has, in other words, the same type.

The situation was further complicated by De Candolle. Having misidentified *Baccharis foetida* L., he also misidentified *Conyza bifrons* L. and transferred that to *Pluchea*. Linnaeus, in Sp. Pl. ed. 1, 861 (1873), based *C. bifrons* upon his earlier polynomial in Hortus Cliffortianus (Hort. Cliff. 405 (1737). In Sp. Pl. ed. 2, 1207 (1763), he referred it to a figure of Plukenet (Pluk., Alm. 141. t. 87. f. 4), and indicated that the plant grew in Europe and in Canada. It was later ascertained that *C. bifrons* was a European *Inula* (incorrectly ascribed to Canada), accordingly to Gray.[22] Gray (ibid.), perceiving both of de Candolle's errors, *viz.* his misidentifications, took up the name *Pluchea bifrons* DC. for the plant of Gronovius and excluded the synonym of Linnaeus (i.e., *Conyza bifrons*).[23] In so doing, apparently to minimize confusion, he dropped the name "*foetida*" entirely and maintained *Pluchea bifrons* of De Candolle, disregarding both types and priority.

Britton, Sterns, and Poggenburg,[24] finding Gray's solution unacceptable, treated the plant as follows: "*Pluchea foetida* (L.) BSP. (*P. bifrons* DC.)." According to present rules, however, De Candolle's misidentification is ignored, and his nomenclatural transfer is accepted, making that of Britton, Sterns, and Poggenburg superfluous.

The specimens at hand indicate that within the range of *Pluchea foetida* (L.) DC., there is occasionally a plant the leaves of which are lanceolate-elliptical, sessile, with cuneate bases, rather than being typically ovate-oblong or oblong-elliptical, with clasping bases. In technical characters these plants do not differ from the general run. It is this leaf-form which was described by Small as *Pluchea tenuifolia*.

There is apparently but a single collection of *Pluchea* in the *P. foetida* complex known from Missouri, i.e., Poplar Bluffs, Aug. 5, 1879, *George W. Letterman* (NY, P, US). The specimens are of immature plants and they were much discolored in drying. It is impossible to ascertain with certainty whether they are

[21] J. J. Dillenius. 1732. Hortus Elthanensis, 106 t. 89, f. 105.

[22] Asa Gray. 1884. Synoptical Flora of N. A. 2 (2): 226.

[23] Gray's notes on the Clayton Herbarium, examined at the Gray Herbarium, courtesy of Miss Marjorie Stone, Librarian, give his interpretation of the Gronovian-Clayton plant as follows: "(Clayton) 451 and 159 = *Eup. fl. albis: Pluchea bifrons!*"

[24] N. L. Britton, E. E. Stearns, and J. F. Poggenburg. 1888. Prelim. Cat. Fl. N. Y., p. 27.

specimens of *P. foetida*, var. *foetida*, or what is described below as *P. rosea* Godfrey. They are cited as doubtfully belonging to the former.

8b. *Pluchea foetida* (L.) DC., var. *imbricata* Kearney, Bull. Torr. Bot. Club **21**: 483 (1894)

P. imbricata (Kearney) Nash, Bull. Torr. Bot. Club **23**: 108 (1896).

Similar to var. *foetida*, but with the uppermost leaves tending to be short triangular-ovate rather than oblong-elliptical; inflorescence having few to numerous heads in tight, rounded clusters at the ends of the branches; heads turbinate-campanulate, *ca.* 9–10 mm. high; phyllaries more closely imbricate, suffused with rose-purplish color, the outer oblong, obtuse-apiculate, grading to linear-acute on the inside. FIGS. 3 & 4, PLATE 20.

Distribution: In habitats similar to var. *foetida*, from southern Georgia to south-central peninsular Florida and in northwestern Florida.

Type Collection: In the original account of this variety, Kearney cited two collections, but did not designate a type. One of those cited, i.e., vicinity of Eustis, Lake Co., Fla., *Nash 1434*, July 16–31, 1894, is here chosen as the type collection because it is represented in several herbaria by characteristic specimens. The specimen in the Herbarium of the New York Botanical Garden is designated as the Type. Isotypes are to be found in C, G, P, US.

Additional specimens examined: **Georgia.** CHATHAM CO.: Thunderbolt, July 21, 1922, *Crawford* (P); COFFEE CO.: wet pine barrens e. of Douglas, *Harper 1430* (CM, G, NY, US); GLYNN CO.: Brunswick, July 26–28, 1895, *Small* (CM, NY); light woods, Jekyl Island, March 26, 1914, *Lee* (NY). **Florida.** ALACHUA CO.: swamp, River Styx near Evinston, Oct. 1, 1940, *Arnold & West* (F); CLAY CO.: near lagoon, Gold Head Branch State Park, near Keystone Heights, *Beck 241* (F); FRANKLIN CO.: moist pine barrens, Apalachicola, *Biltmore Herbarium 2956a* (G, NY, US); HAMILTON CO.: cypress pond, 6 miles e. of Jasper, July 26, 1946, *Arnold & West* (F); HIGHLANDS CO.: Istokpoga Prairie, *Small et al. 11517* (G, NY, US); swamp, Carter Creek, e. of Sebring, May 16, 1948, *Garrett* (F); LAKE CO.: peat prairie west of Clermont, July 10, 1935, *Davis* (F); LEE CO.: ditches, *Hitchcock 162* (CM, G, NY, US); MARION CO.: edge of pond by Lake Kerr, 12 miles e. of Eureka, Sept. 24, 1941, *West & Arnold* (F); ORANGE CO.: Clarcona, Aug. 23, 1899, *Meislahn* (US); PALM BEACH CO.: glade w. of West Palm Beach, Aug. 10, 1911, *Harshberger* (P); POLK CO.: edge of cypress swamp, e. of Dundee, *McFarlin 6217* (F); ST. JOHNS CO.: cypress pond w. of Crescent Beach, Oct. 14, 1941, *West & Arnold* (F); ST. LUCIE CO.: pineland, e. of Okeechobee, June 19, 1948, *Ledin* (F); TAYLOR CO.: 14 miles s. of Perry, Oct. 8, 1940, *Murrill* (F).

The characters by which this variety is distinguished from var. *foetida* tend to grade imperceptibly into those of the latter. However, specimens exhibiting the distinguishing characteristics are localized with respect to the range of the whole population of the species. It is, therefore, recognized as of geographic varietal rank as it was originally described. In this connection several facts lead to the conclusion that Nash, when he raised this taxon to specific rank, was, in his discussion of its change of status, comparing its characteristics not with the plant shown to be *P. foetida*, but with the rose-flowered one which was discussed earlier in relation to *P. foetida* and which is described below as *P. rosea* Godfrey. Each of the characteristics which he attributes to *P. foetida* applies much better

to *P. rosea* than to *P. foetida*, var. *foetida* as delimited above. This conclusion is borne out by an examination of other specimens collected by Nash (and labelled *P. foetida*—presumably by him) in the identical vicinity and at the identical time that he collected his number *1434* on which var. *imbricata* was in part based. Indeed, one of these Nash specimens has been chosen as the type of *P. rosea*.

That Kearney, or Nash, working in Florida, should take the rose-flowered plant to be *P. foetida* is not surprising; it is much the more common there. Moreover, it is the only one of the group occurring in southernmost Florida. It may be recalled that Urban made the same error when he described *P. Eggersii*. His collections show that he was familiar with the rose-flowered plant in the Bahama Islands which was then passing, and has up to now passed, as *P. bifrons* or *P. foetida*. When he discovered Egger's collection of the white-flowered form from the Dominican Republic, hitherto unknown in the West Indies, he gave to it a new name. It is singularly interesting that Kearney, who happened to be working at the extreme southern edge of the range of *P. foetida* in Florida, did describe a plant which appears to be varietally distinct from the typical, even though he, and later Nash, may have been mistaken about the identity of the plant from which they thought they were segregating it.

9. **Pluchea rosea** Godfrey, sp. nov.

Perennis, herbis cano-viridis, odoris; radicibus ex brevibus ramis rhizomatis fibris; caulibus erectis, 3–8 dm. altis, frequentissime aliquot ex singularo rhizomato emergeris; caulibus imis glabratis; superficibus cauliorum foliorum involucrorumque cum sessilibus hemisphaericis glandulis et resinosis atomis, caulibus superioribus pubescentibus, ramis inflorescentiorum involucrisque glandularis tomentosisque, aliquando rufescentibus; foliis inferioribus mediisque sparsim pubescentibus, superioribus copiose pubescentibus vel tomentosis, aliquando rufescentibus;—aut caulibus foliis involucrisque non pubescentibus aut solum sparsim pubescentibus cum piliis glandulosis et usque scabris cum rigidis pappilosis aut spiculatis glandulis; foliis sessilibus, imis oblanceolatis basis truncatis vel cuneato-truncatis; foliis mediis majoribus latioribusque, oblongis vel ovato-oblongis, elliptico-oblongis basis cordato-subamplexicaulibus, aut lanceolatis vel oblanceolatis basis cuneato-truncatis, ca. 2–7 dm. longis, 0.5–3 cm. latis; foliis superioribus minoribus sed similibus; margis foliorum apiculato-serratis; inflorescentibus capitulis multis vel paucis, paniculato-cymosis dispositis; involucris campanulatis, turbinato-campanulatis, aut turbinatis, 4–7 mm. altis, 3–6 mm. latis; phyllaribus pluribus seriis dispositis, exterioribus ovato-lanceolatis vel lanceolatis, acuminatis, mediis oblongis, obtusis vel acuminatis, interioribus linearibus, acutis vel attenuatis; phyllaribus plus minusve roseis; pappi seteis 3.5–4.5 mm. longis; achaeniis ca. 1 mm. longis, rubrofuscis, 6-nervis, nervis hirsutis; corollis purpureo-roseis.

Distribution: Savannas, flatwoods, pond margins and ditches, generally in habitats which have a markedly fluctuating water table, thus alternately very wet to dry; outer coastal plain from eastern North Carolina through Georgia, throughout Florida, westward to eastern Texas, in the states of San Luis Potosí and Quintana Roo, Mexico, and in the Bahama Islands and Cuba.

9a. **Pluchea rosea** Godfrey, var. **rosea.**

Plants 3–5.5 dm. high; stems glabrate below; surfaces of the stem, leaves, and involucres with sessile, hemispherical glands and resinous atoms, upper stems pubescent, the inflorescence branches and involucres tomentose, sometimes rufescent; lower and median leaves sparsely pubescent, the upper copiously pubescent to tomentose, typically tomentose, sometimes rufescent; leaves sessile, the lower oblanceolate with truncate to cuneate-truncate bases; median leaves larger and wider, oblong to ovate-oblong, elliptical-oblong with cordate subclasping bases, or lanceolate to oblanceolate with cuneate-truncate bases, about 2–7 cm. long, 0.5–3 cm. wide; upper leaves smaller but similar; margins of the leaves apiculate-serrate; inflorescence with many to few heads paniculately cymosely disposed; involucres campanulate, turbinate-campanulate, or turbinate, 4–6 mm. high; the outer phyllaries from ovate-lanceolate, short-acuminate to lanceolate, long-acuminate, and foliaceous; median phyllaries from oblong or oblong-lanceolate obtuse or short-acuminate to lanceolate and acuminate, the inner linear, acute to attenuate; phyllaries more or less rose-purplish; pappus setae 3.5 to 4 mm. long; corollas rose-purplish. FIGS. 5 & 6, PLATE 21.

Distribution: As given above for the species, with the exception of the portion of the population occurring in Mexico.

Type Specimen: **Florida:** lake shores, vicinity of Eustis, Lake Co., May 16–31, 1894, *Nash 758* (G); Isotypes: C, CM, NY, US.

Some additional specimens examined: **United States. North Carolina.** BRUNSWICK CO.: sphagnous, exsiccated pool in wire-grass savanna, near Inland Waterway along Caswell Beach Road, Aug. 20, 1948, *Godfrey 48408* (C, F, G, GA, NCS, SM); CARTERET CO.: abundant around the margin of a very small sink-hole pond in sandhills, w. of Morehead City, Aug. 6, 1949, *Godfrey 49825* (G, NCS, SM); CRAVEN CO.: wet, black swamp muck in edge of cypress pond, ½ mile s. of intersection of U. S. Rt. 70 and N. C. Rt. 101, Aug. 11, 1939, *Quarterman 315* (D); DARE CO.: bog, 2 miles s. of Manteo on N. C. Rt. 345, Roanoke Island, July 23, 1949, *Radford 4646a* (NC). **South Carolina.** BERKELEY CO.: burnt-over pineland, near Appia, Aug. 9, 1939, *Martin 1184* (D, NA); CLARENDON CO.: pine barrens, 1 mile s. of Manning, May 30, 1914, *Stone 540* (P); DARLINGTON CO.: pine flat about 1 mile e. of Kellytown, July 19, 1941, *Smith 845* (NC); GEORGETOWN CO.: abundant in savanna, 4.5 miles w. of Georgetown, Aug. 21, 1950, *Godfrey & Boyce 50436* (CM, D, F, GA, NA, NCS, NY, SM, T); HORRY CO.: low, damp, sandy woods back of Myrtle Beach, July 2, 1927, *Coker & Holland* (NC); ORANGEBURG CO.: Eutawville, Sept. 6–11, 1909, *Eggleston 4996* (G, NY); SUMTER CO.: dry savanna, Aug. 5, 1884, *Smith* (US). **Georgia.** BAKER CO.: in moist soil of dry bed of Mossy Pond, s.w. corner of Baker Co., Aug. 29, 1941, *Duncan 4054* (GA); CANDLER CO.: moist sandy meadow near Upper Lott's Creek Church, 10 miles n.e. of Metter, July 4, 1936, *Pyron & McVaugh 744* (GA); flat land, about 5 miles s.w. of Folkston, July 22, 1937, *McKellar* (GA, US); CHATHAM CO.: low meadow, 5 miles s.w. of Savannah, July 21, 1927, *Wiegand & Manning* 3291 (G); CLINCH CO.: abundant in wet drainage ditch at roadside, 8 miles s. of Homerville, Oct. 15, 1950, *Godfrey 50816* (C, CM, D, GA, NA, NCS, SM); GLYNN CO.: Jekyl Island, June 1923, *McAtee 3258* (US); JENKINS CO.: low, open pinelands, sandy soil 7.5 miles s. of Millen, flowers bright lavender, July 8, 1949, *Duncan 3904* (GA); MCINTOSH CO.: in water on edge of marsh near Darien, June 20, 1936, *Correll 5458* (D, NA); SCREVEN CO.: sandy, grit region 10 miles w. of Sylvania, Aug. 7, 1942, *Duncan 5566* (GA); SUMTER CO.: margin of pine-barren pond, w. of Leslie, Aug. 31, 1900, *Harper 554* (CM, NY, US); TIFT CO.: App. For. Exp. Sta., 8 miles e. of Tifton, Aug. 26, 1941, *Shepherd 187* (GA); WARE CO.: roadside ditch in intermediate pine barrens, Suwannee Lake, fls. light purple,

leaves viscid, July 4, 1931, *Harper 250* (P, NY); WAYNE CO.: flat, wet pinelands, s.w. of Jesup, Aug. 27, 1941, *Duncan 3994* (GA, SM). **Florida.** ALACHUA CO.: open swamp, flatwoods, 4 miles e. of Silver Springs, vicinity of Gainesville, July 16, 1939, *Murrill* (NCS); BAKER CO.: ditch near flatwoods pond, 9 miles s. of MacClenny, July 22, 1946, *West & Arnold* (F); BAY CO.: sandy soil, Lynn Haven, Oct. 15, 1921, *Billington* (US); BRADFORD CO.: flatwoods ditch, n.e. of Brooker, Sept. 12, 1946, *Dennison & Arnold* (F); CHARLOTTE CO.: edge of flatwoods pond, May 14, 1946, *Frye* (F); CITRUS CO.: bushy flatwoods, Rd. 15, Red Level, June 6, 1941, *Murrill* (F); COLLIER CO.: open sandy woods, Naples, Aug. 13, 1933, *Degener 4948* (NY); COLUMBIA CO.: flatwoods pond, Lake City, July 26, 1898, *Rolfs 962* (CM); DIXIE CO.: hammock, 3.7 miles n.e. of Cross City, Aug. 16, 1937, *"Pasture Survey"* (F); DUVAL CO.: moist open pine barrens near Jacksonville, June 30, 1894, *Curtiss 5039* (C, CM, G, NCS, NY, US); FRANKLIN CO.: low, moist, open soil, St. Vincent Island, June 25, 1936, *Correll 5580* (D, NA, NCS); GULF CO.: waste places, Apalachicola, *Saurman* (P); HIGHLANDS CO.: creek, hammock border, Lake Jackson-Lake Sebring, Sebring, June 1, 1948, *Garrett* (GA); JACKSON CO.: Grand Ridge, June 9, 1936, *Knight* (F); LAKE CO.: vicinity of Eustis, June & July 1894, *Hitchcock 1045* (CM, F); LEE CO.: cypress and pine land, vicinity of Fort Myers, Apr. 3, 1916, *Standley 122* (C, G, NY, US); Sanibel Island, May 17, 1901, *Tracy 7254* (CM, G, NY); LEON CO.: near Tallahassee, *Berg* (NY); MANATEE CO.: Braidentown, *Tracy 7424* (CM, G, NY, US); border of hammock, Braidentown, June 14, 1916, *Cuthbert 1317* (F); MARION CO.: open swamp in flatwoods, 4 miles e. of Silver Springs, July 16, 1939, *Murrill 682* (US); OKEECHOBEE CO.: cypress pond, 4 miles n. of Okeechobee, Apr. 23, 1946, *West & West* (F); ORANGE CO.: Lake Brantley, July 11, 1894, *Lewton* (CM, NY); PALM BEACH CO.: in a *Taxodium ascendens* swamp along Road 25, about 31 miles e. of Belle Glade, Feb. 23, 1941, *Deam 60831* (D); PINELLAS CO.: pinelands, Lake Butler, Apr. 23, 1927, *O'Neill* (NA); POLK CO.: sandy-peaty pond margin, just n.e. of Davenport, Oct. 17, 1950, *Godfrey 50892* (C, NCS); PUTMAN CO.: edge of Interlachen Lake, July 2, 1939, *Murrill 722* (US); ST. JOHNS CO.: salt marsh, Crescent Beach, Oct. 14, 1941, *West & Arnold* (F); UNION CO.: cypress bog, Lake Butler, Aug. 18, 1939, *Murrill* (F); VOLUSIA CO.: margin of small, shallow pond, 3.5 miles s. of Orange City, *Godfrey 50882* (C, CM, D, NA, NCS, SM, T). **Alabama.** Without locality: July 1840, *Buckley* (P); GENEVA CO.: 2 miles n.e. of Sampson, July 5, 1940, *Campbell 60* (D); MOBILE CO.: wet ground, Spring Hill, Aug. 1919, *Graves 1211* (US). **Mississippi.** Deer Island, Aug. 25, 1891, *Seymour 91825.22* (D, G, NC, SM); Deer Island, *Earle* (D); HARRISON CO.: Biloxi, July 25, 1896, *Pollard 1004* (NY, US); Biloxi, July 4, 1897, *Tracy 3586* (NY); Biloxi, July 16, 1898, *Tracy 4365* (US); Biloxi, Sept. 6, 1900, *Tracy* (G); JACKSON CO.: Ocean Springs, July 25, 1896, *Pollard 1004* (G); Ocean Springs, 1889, *Earle* (D). **Louisiana.** SAINT TAMMANY PARISH: vicinity of Covington, *Bro. Arsène 11402* (G). **Texas.** HARDIN CO.: infrequent in moist sandy soils, 2.5 miles w. of Silsbee, Oct. 2, 1945, *Cory 49900* (SM); in low pineland, near Ariola, fls. rose-pink, Aug. 18, 1942, *Lundell & Lundell 11499* (SM); 3.5 miles n. of Camp Jackson, Sept. 12, 1946, *Cory 19697* (NA); in wet pineland, about 4 miles n. of Voth, off highway 69, fls. pinkish, July 18, 1945, *Lundell 14070* (SM); HARRIS CO.: in low coastal prairie, between Houston and Conroe, fls. pinkish, July 19, 1944, *Lundell & Lundell 13104* (G, SM).

Bahama Islands. GREAT BAHAMA: palmetto lands, Barnett's Point, Feb. 5–13, 1905, *Britton & Millspaugh 2652* (CM, NY); ELEUTHERA: marsh, Glass Window to Harbor Island, Feb. 17, 1907, *Britton & Millspaugh 5392* (CM, NY); NEW PROVIDENCE; Hog Island, in paludosis, Feb. 18, 1888, *Eggers 4103* (DN); moist, sandy places, Hog Island, May 30, 1909, *Wilson 8266* (CM, NY); *Cooper 76* (CM, G, NY); fresh water swamp, S. W. Bay, Sept. 3, 1904, *Britton & Brace 517* (CM, NY); ANDROS: swampy ground, Nicholl's Town and vicinity, Mar. 13–15, *Brace 6862* (CM, NY); savannah, near Staniard Creek, Northern Section, heads purplish, Feb. 1–3, 1910, *Small & Carter 8893* (CM, NY); Red Bays, Apr. 17, 1890, *Northrop & Northrop 498* (CM, G, NY). **Cuba.** ISLE OF PINES: Lieux secs, May 1–4, 1944, *Marie-Victorin & Alain 11* (G); PINAR DEL RÍO: Herradura, Sept. 18, 1888, *Hermann 924* (NY); HAVANA: El Rosario, Mar. 1926, *Juzepczuk 4088* (US); common in saline or brackish sedge-swales, Cayo Largo, Apr. 4–9, 1948, *Proctor 3158* (P); LAS VILLAS: Sabana de Motembo, Aug. 9–11, 1920, *Bro. Léon & A. Loustalot 9419* (CM).

9b. *Pluchea rosea* Godfrey, var. **mexicana** Godfrey, var. nov.

Ex characteribus maximam partem var. *rosea* similis sed planta altiora, ad 8 dm. alta; multo minus glanduloso-pubescentibus usque, sed pappiloso-glandulosis vel spiculati-glandulosis usque, non tomentosis; phyllaribus omnibus lanceolato-acuminatis vel lineari-acuminatis, conspicue purpureo-roseis; involucris conspicue turbinato-campanulatis, 6–7 mm. altis. FIG. 11, PLATE 22.

Distribution: Savannas, alkaline plains, San Luis Potosí, and Quintana Roo, Yucatán Peninsula, Mexico.

Type Specimen: SAN LUIS POTOSÍ: by streams, alkaline plains, Hacienda de Angostura, Aug. 5, 1891, *Pringle 3813* (G); Isotypes: C, CM, NY, P, US.

Additional specimens examined: SAN LUIS POTOSÍ; Media Luna, near Rio Verde, *Palmer 75* (CM, G, NY, US); Minas de San Rafael, Angostura, June, 1911, *Purpus* (C, CM, G, NY, US); Minas de San Rafael, plains between Tiburcio and Angostura, July, 1911, *Purpus* (CM, G, NY, US); QUINTANA ROO: in savannah, Lake Chichancanab (Laguna Chankabnab), *Steere 2407* (CM, US).

In the foregoing general discussion of the identity of *Pluchea foetida* and its allies, the significant point was made that the taxa here treated as *P. rosea*, var. *rosea* and *P. rosea*, var. *mexicana* were hitherto identified with the name *P. foetida* (L.) DC., the true basis for that name was established and the population identifiable with it was delimited. The results of an analysis of the remainder of the population formerly confused with *P. foetida*, and the naming and delimitation of its elements, is accomplished just above. No further discussion of that problem is deemed necessary.

The portion of the population of *Pluchea rosea* which occurs in Mexico is geographically disjunct from the remainder. Morphologically, the Mexican material is, in general, very similar to the plant of the southeastern United States and the West Indies, but the copious pubescence characteristic of the leaves, stems, and involucres of the latter plant is essentially absent. Rather, these parts of the Mexican plants are rendered very scabrous with papillose or spiculate glandular protuberances quite unlike anything found in these plants elsewhere. Also, the heads are a little larger, and all the outer and median phyllaries of the involucres are consistently lanceolate-acuminate rather than varying from ovate-lanceolate to oblong or to oblong-lanceolate as in the others. For these reasons varietal segregation into two taxa is proposed.

AN EXCLUDED SPECIES

Pluchea auriculata Hemsley, Diagn. Pl. Nov. Mex. et Centr.-Amer. 32 (1879)

Type Specimen: MEXICO: Guanajuato, *Hartwig 112* (K); isotype, (G).

An examination of the type specimen of this putative species of *Pluchea*, which was made available through the courtesy of the staff of the Royal Botanic Gardens, Kew, revealed that the plant was incorrectly placed in *Pluchea*. The heads do not have flowers with corollas of the two kinds characteristic of this genus; rather the flowers all have corollas which are filiform-tubular and truncate at the apex. It is apparently a specimen of some species of *Conyza*.

A NEW SOUTH AMERICAN SPECIES

Pluchea microcephala Godfrey, sp. nov.

Perennis, basis caudicibus duris lignis, superioribus herbaceis, 0.5–2 dm. altis; caudicibus ex basis decurrentis foliorum alatis; imis caudicibus glabratis, plus minusve glaucis, supra exigue inaequalibeque canescentibus; foliis basis decurrentibus, imis alato-petiolis, glabratis, mediis superioribusque sessilibus, oblanceolatis vel lanceolatis aut lineari-lanceolatis, apicibus obtusis vel acutis, margis serrulatis, glabratis vel exigue inaequalibeque canescentibus; inflorescentium cum multis parvulis capitulis paniculato-cymosis dispositis; capitulis anguste campanulatis, ca. 6 mm. altis, 3 mm. latis; phyllaribus 4–5 seriis dispositis, glabris, tenuis, stramineis, exterioribus ovatis, obtusis, mediis oblongis, obtusis, interioribus linearis, acutis vel attenuatis; corollis purpureo-roseis; achaeniis ca. 0.8 mm. longis, fuscis, glabris, 6-nervis, nervis membranaceis. FIGS. 18 & 19, PLATE 23.

Distribution: Northwestern Argentina, and northeastern Peru.

Type Specimen: ARGENTINA: al N. O. a 6 km. Pirane, Pirane, Formosa, Dec. 5, 1945, *T. Morel 518* (CM:1299928).

Additional specimens examined: **Argentina.** JUJUY: perennial herb 0.5–1 m., single or branched stems at the base, fl. heads pinkish purple, in river bottom, soft mud below river bank, partially exposed to sunlight, alt. 800 m., Río Lavagin, 15 km. southeast of San Pedro de Jujuy, Depto. San Pedro, Oct. 11, 1938, *Eyerdam and Beetle 22535* (G); SALTA: El Tabacal, Dep. Oran, Feb. 17, 1943, *T. Myer 4519* (C); **Peru.** CHICLAYO: low bush usually below 0.5 m. to 2 m. along irrigation ditches, fl. purple, common in the plains about Chiclayo, in dry plains, grows strongly where it gets drainage water, Depto. Lambayeque, April 13, 1939, *Stork 11429* (C).

The winged stem and foliage of this species suggest *Pluchea suaveolens*. It is clearly distinguished from that plant, however, by its much smaller heads which are narrowly campanulate rather than broadly hemispherical, by its thinner, smaller, glabrous, differently shaped and more stramineous phyllaries, by its much more characteristically and conspicuously rose-purplish corollas, by the absence of glandularity throughout, and by the entirely different character of the pubescence on the stems and leaves.

SUMMARY

This paper consists of a taxonomic revision of the species in the section *Stylimnus* of the genus *Pluchea* (Compositae) occurring in North America, Central America, and the West Indies. There are 13 taxa treated, there being nine species, each of four of which has one geographical variety. An artificial diagnostic key to the taxa is presented followed by a systematic treatment of each taxon, including synonymy, a description, and detailed citations of specimens.

The specimen citations represent documentary evidence both of the material upon which the descriptions of the taxa are based and of the geographical distribution of each. Approximately 3,000 specimens from 17 herbaria were examined in the course of the study.

One taxon hitherto regarded as of varietal rank is raised to specific rank. One species and a variety of it are described as new to science.

A new South American species, discovered incidental to the study of the North American Plucheas, is also described.

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EXPLANATION OF PLATES

(Plates arranged by Dr. Carroll E. Wood, Jr.)

PLATE 20

FIG. 1. *Pluchea foetida* (L.) DC. Habit sketch of uppermost portion of plant. ×0.6

FIG. 2. *P. foetida* (L.) DC. Heads. ×1.2

FIG. 3. *P. foetida*, var. *imbricata* Kearney. Habit sketch of uppermost portion of plant. ×0.6

FIG. 4. *P. foetida*, var. *imbricata* Kearney. Heads. ×1.2

PLATE 21

FIG. 5. *Pluchea rosea* Godfrey. Habit sketch of uppermost portion of plant. ×0.67

FIG. 6. *P. rosea* Godfrey. Mature heads. ×1.3

FIG. 7. *P. longifolia* Nash. Habit sketch of uppermost portion of plant. ×0.5

FIG. 8. *P. longifolia* Nash. Heads. ×1.3

PLATE 22

FIG. 9. *Pluchea purpurascens* (Sw.) DC., var. *succulenta* Fern. Heads. ×1.2

FIG. 10. *P. odorata* (L.) Cass. Heads. ×1.6

FIG. 11. *P. rosea* Godfrey, var. *mexicana* Godfrey. Heads. ×2

FIG. 12. *P. salicifolia* (Miller) Blake, var. *canescens* (Gray) Blake. Heads. ×1.6

FIG. 13. *P. parvifolia* (Gray) Godfrey. Heads. ×2

FIG. 14. *P. suaveolens* (Vell.) Kuntze. Heads. ×0.8

FIG. 15. *P. salicifolia* (Miller) Blake. Heads. ×2

FIG. 16. *P. purpurascens* (Sw.) DC. Heads. ×1.6

FIG. 17. *P. camphorata* (L.) DC. Heads. ×2.4

PLATE 23

FIG. 18. *Pluchea microcephala* Godfrey. Habit sketch showing uppermost portion of plant. ×0.9

FIG. 19. *P. microcephala* Godfrey. Heads. ×3.8

PLATES

PLATE 20

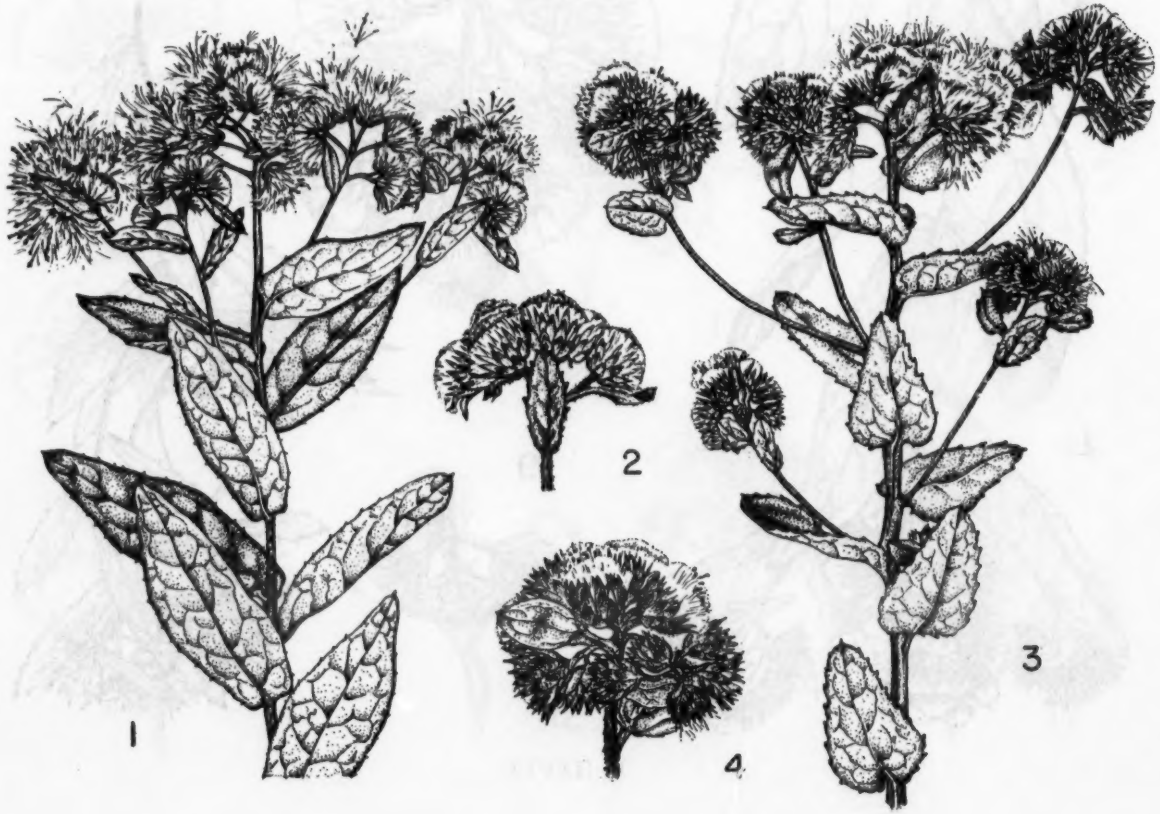

PLATE 21

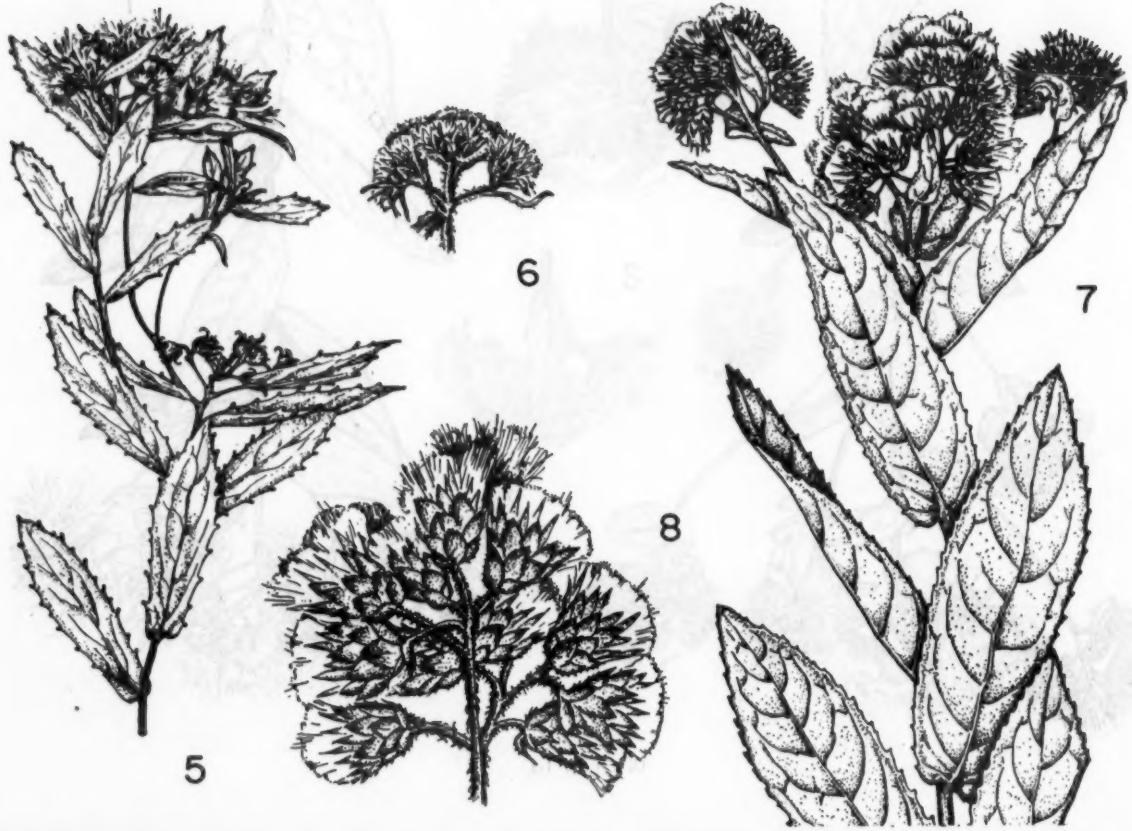

PLATE 22

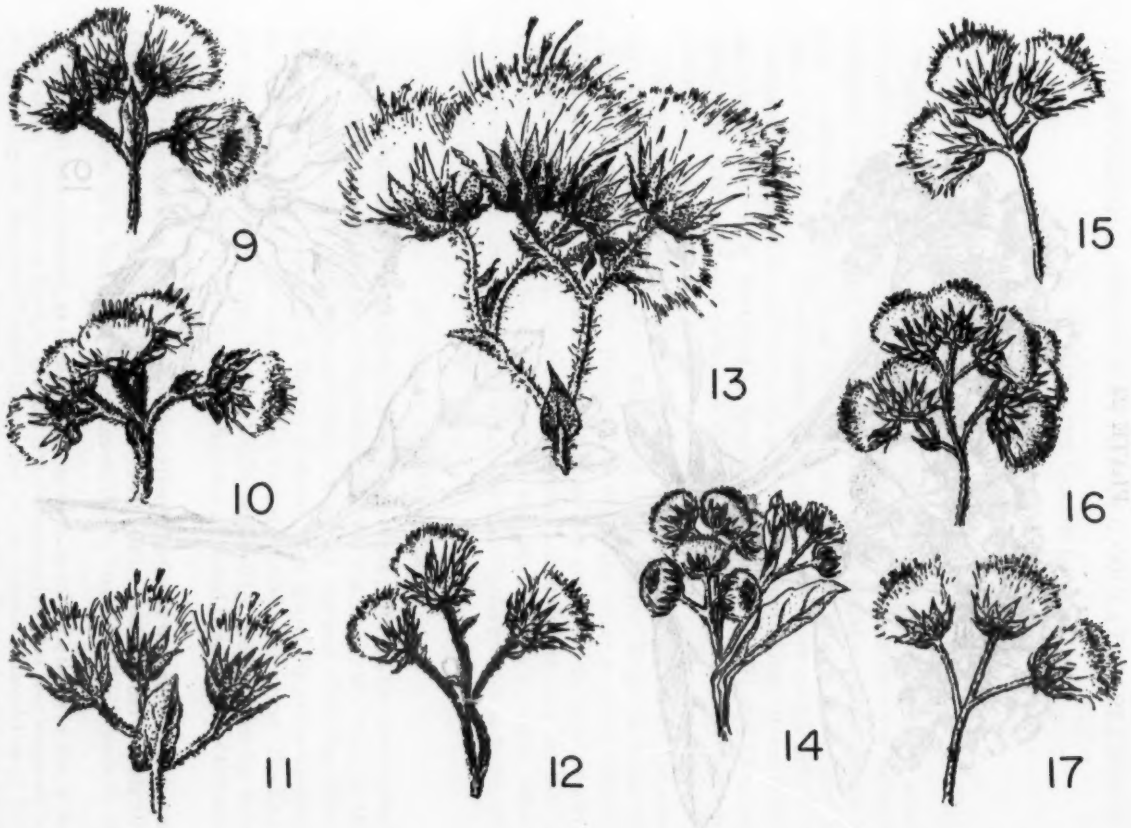

PLATE 23

THE SPORANGIAL PHASE OF *PYTHIUM UNDULATUM* PETERSEN[1]

By E. Kathleen Goldie-Smith

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Plates 24–26

Pythium undulatum appears at once as an outstanding member of the genus on account of its large ellipsoid sporangia and the general coarseness of its habit compared with most other species. In its mode of sporulation also it seems to show a striking departure from the usual concept of the process in *Pythium*, in that a high degree of differentiation is reached by the protoplasm before its emergence into the vesicle at the mouth of the sporangium. While other interesting aspects have been discussed at length by Dissmann (1927) and Drechsler (1946), the process of sporulation in this species has not previously been described in detail.

The present study was begun in October, 1946, and carried out for the most part at Royal Holloway College, Englefield Green, Surrey, England, under the direction of Miss Elizabeth M. Blackwell, to whom the writer is deeply indebted for her painstaking and invaluable help. It has been continued at the University of North Carolina, U.S.A., where most grateful acknowledgements are due to Dr. John N. Couch for his encouragement and advice. Among many others who helped by discussion and criticism, particular thanks are due to those who translated parts of the references.

Material and Methods

Three isolates have been studied in pure single-spore culture. One was obtained from decaying waterlily leaves (*Nymphaea tuberosa* var. *rosea*) in a pond in the botany garden at Royal Holloway College. A second was sent with some drawings by Miss van Beverwijk of Baarn, Holland, here gratefully acknowledged. Both of these isolates were brought to the United States by the writer. The English one was subsequently lost, but was kindly replaced by another from the same source by Miss B. M. Brady. A third isolate was obtained from soil collected near Highlands, North Carolina, U.S.A.

Pythium undulatum has been found on various species of waterlily by Petersen (1910), Dissmann (1927), and Drechsler (1941, 1946). Petersen did not ascribe any parasitism to it, but made the comment that it "seems to live especially on

[1] This work was submitted, as far as it had progressed, as part of a thesis for the M. Sc. degree of the University of London, in June, 1948. Subsequently, it has been supported by the Rose Sidgwick Memorial Fellowship awarded by the American Association of University Women (through the British Federation of University Women) for the year 1948–9 at the University of North Carolina, and by the Col. and Mrs. John S. Sewell Scholarship awarded by the Highlands Biological Station for a period of study there in the summer of 1950.

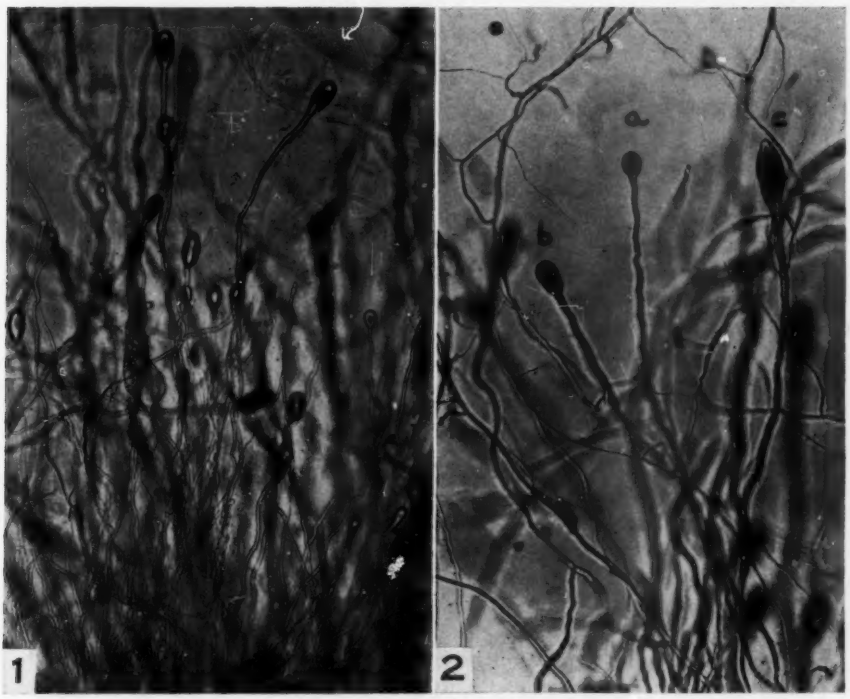
decaying leaves and stems of *Nuphar* and *Nymphaea*," and that he had occasionally found it "on old fruits of Iris and on branches of trees." Drechsler, on the other hand, relates the premature discoloration of the waterlily leaves to attack by this fungus. At Royal Holloway College, however, repeated attempts to reproduce this discoloration in the laboratory by inoculating healthy leaves with a pure culture of *P. undulatum* met with no success, although a darkening of the leaf around an inoculum of another *Pythium* species from the same source occurred in twenty-four hours or less. These observations, and the occurrence of *P. undulatum* in a number of ponds in the College grounds where there are no waterlilies, together with its ready saprophytic growth on many other substrates such as boiled rose hips, twigs, and hemp seeds, suggest that, if this species is a parasite at all on these waterlilies, it is a very weak one.

A supply of sporangia for study was obtained by a method, described in full in a previous paper (Goldie Smith, 1950), in which sterile halved hemp seeds are placed on the growing margin of a strong clean culture on cornmeal agar, and transferred after a number of hours to sterile water. The first stage of this method can be carried out at any laboratory temperature, the length of the time that the seeds may be left on the agar varying between ten and twenty hours, the higher temperatures necessitating the shorter times. For the second stage, however, when the seeds are in water, the temperature must be low, and the best results have been obtained at 18–20°C. At temperatures above and below this, most of the vigor of the mycelium goes into the formation of vegetative hyphae rather than of sporangia. The effect of the age of the mycelium, as well as the constitution of the medium in which it is grown, on the time required for the development of sporangia after transference to water, was observed by Dissmann in his extensive physiological studies on *P. undulatum*. The time also varies among the strains, which have different growth rates. The supply of sporangia from a single culture can be kept up for some time by continual irrigation or by repeatedly refreshing the water, but it was found more expedient in this study to plan for a fresh supply each day.

The Form and Development of the Sporangium

The variation in the size and shape of the sporangia is one of the most remarkable features of this species. In a vigorous culture, the sporangium is ellipsoidal, often 130 x 35μ in size under favorable conditions, occasionally reaching a length of 170μ. The largest sporangia are obtained on natural media such as decaying waterlily leaf, or boiled rose hips, and a good size is reached on hemp seeds when the method described above is used. Large vigorous sporangia are formed when portions of a culture on waterlily decoction agar or hemp seed agar are put into distilled water which is repeatedly refreshed. The sporangium is borne on a sporangiophore 6–8μ wide, which may widen to 12–15μ immediately under the sporangium. This widening is a very distinctive feature. The sporangiophores are strongly aerobic, rising vertically from thick stolon-like hyphae close against the seed, which also bear the more delicate vegetative hyphae, 3–5μ in width, that become profusely branched and eventually form a dense mycelial

weft around the substratum (Fig. 40–41). The primary sporangiophores are usually unbranched and loosely (Fig. 1) or closely (Fig. 39) undulating. They give the appearance under the microscope of coarse untidy hair (Fig. 1). With chlor-iodide of zinc they give a pronounced violet-pink color. As the culture declines in vigor, or if the nature of the media and other conditions such as aeration and temperature are less favorable, smaller sporangia of entirely different



FIGURES 1–2. Developing sporangia of *Pythium undulatum* Petersen. FIG. 1. Early stages, mostly before the formation of the septum. ×80. FIG. 2. Three successive stages, a, b, and c, respectively. ×120.

proportions are produced. These are ovoid or obpyriform instead of ellipsoid (Fig. 26, 28, 31), of such dimensions as 50 x 30μ, usually conspicuously papillate (Fig. 54), and often without any widening of the sporangiophore immediately below. They are formed both on branches of the thick sporangiophores (Fig. 36) and on the narrow hyphae of the mycelial weft which, under conditions which are less favorable for the large sporangia, is profuse from the first. During sporulation, these smaller sporangia go through the same process as the large ones. This change in proportions is particularly interesting in view of the fact that the ratio of length to breadth is generally found more or less characteristic for a given

species, as stated for *Phytophthora* by Miss Blackwell (1949, p. 11). Dissmann carried out experiments with solutions of different composition and concentration, and came to the conclusion that while the size of the sporangia is variable, the largest being formed in pondwater to which waterlily leaf decoction had been added, their shape is fairly constant. Some of the sporangia formed by germinating chlamydospores figured by Drechsler (1946, p. 854) are of the small ovoid or obpyriform type, but he has made no comment on the fact. Sparrow (1932) noted a great diversity of shapes, but his observations refer to cultures "under foul environmental conditions", when, indeed, a variety of abnormalities may be induced, as described in the following section. A similar response to external conditions is not uncommon in the genus *Pythium* and may also be observed in *Phytophthora*, but is generally much less marked than in *Pythium undulatum*. In this species, a unique type of dimorphism may be observed in a single culture, when the thick, sparsely- or un-branched sporangiophores bearing the large, empty first-formed sporangia are found passing through a weft of much-branched, finer hyphae bearing sporangia of entirely different proportions.

Whatever the ultimate shape of the sporangium, it originates as a subspherical terminal swelling. It remains subspherical until it has reached a considerable size, after which, in a vigorous culture, it develops into an elongated shape with a blunt apex. When the swelling has attained its maximum size, it is cut off from the sporangiophore by a septum, the plane of which is usually flat at first, but often later becomes arched up into the sporangium. After its formation, the apex of the sporangium loses its blunt outline and becomes more pointed, and the sporangium assumes its characteristic ellipsoid shape, with its greatest width about one-third of its length behind the apex. Early stages are shown in figure 1, and three stages, represented successively by a, b, and c, in figure 2. The protoplasm of the mature sporangium has a coarse dark appearance, owing to the presence of numerous, relatively large oildrops (Fig. 53).

SPORULATION

The first sign that a sporangium is preparing for discharge is in the apex, which gradually becomes pale as the granular contents withdraw from it until there is a narrow completely hyaline zone separating the granular protoplasm from the tip of the sporangium, where the wall is slightly more refractive than that of the rest of the sporangium (Fig. 15). The brightness of this region increases as the refractive part of the wall thickens and its area extends forming a gleaming cap over the more or less pointed apex, with a slightly convex thickening directed inwards from the very tip (Fig. 16). Viewed in optical section, the zone between this cap and the protoplasm is completely clear, while at high and low levels of focus some granules may be seen extending right up to the cap; thus it appears that the clear zone is an almost spherical region lying in a cuplike depression in the protoplasm. This zone corresponds with the "subapicular vacuole" described in other species by Butler (1907). At this time, numerous small spherical hyaline regions are clearly seen scattered throughout the protoplasm, which now appears less coarsely granular than before (Fig. 6). When a dilute solution of neutral red

is applied as a vital stain at this stage, these vacuoles color only very slightly. They are not contractile, and are fairly constant in position. The sporangium gradually assumes a remarkable and characteristic appearance due to their presence and the "lumping" (*vide* Butler, 1907) of the protoplasm around them. The limits of any particular mass cannot be exactly distinguished, and the effect is most striking under low rather than high magnification (Fig. 7, 15).

The cap begins to lose its glistening appearance and becomes grey and translucent, though still remaining rather refractive; and the subapicular region gradually becomes re-occupied by the granular protoplasm, which then extends again up to the hyaline cap (Fig. 11, 12). The equidistant vacuoles become very bright and conspicuous. Many of them are so superficial as to appear in surface view as distinct, regularly-spaced circular holes, and in side view as shallow, cuplike depressions in the protoplasm. Deeper indentations of the protoplasm become visible at the apex and around the periphery, as the separation of the spore masses becomes more distinct, and narrow cracks and splits, which have been seen colored with neutral red, develop throughout the protoplasm (Fig. 16). Sometimes, differentiation proceeds no further than this before discharge. Frequently, however, the spores become still more clearly outlined as polygonal blocks, and for a number of seconds appear in zig-zag formation down the middle of the sporangium. Figures 11 and 12 are successive pictures of the same sporangium taken just before discharge, with a slightly higher level of focus in figure 12. A third picture was taken, but the image was blurred by the outflow of the protoplasm which occurred at the same time, demonstrating that this sporangium was indeed ready for immediate discharge. At discharge, the vacuoles are less conspicuous, and the spores are closely pressed together.

The hyaline cap becomes softer and greyer, swelling into a thick layer and no longer appearing to be separated from the underlying protoplasm by a sharp line. As in *Phytophthora* (Blackwell, 1949, p. 11), this soft part of the papilla seems to be derived from the inner part of the apical wall, probably by the action of the underlying protoplasm upon it. It is readily stainable at this stage, and remains as a deeply-colored cap when the protoplasm is withdrawn from it by plasmolysis. The protoplasm immediately under it may also be deeply stained. Sparrow (1931, a) believes that in *P. dictyosporum* it is from this protoplasmic region that the papilla is derived.

The cap swells into a thick hyaline drop (Fig. 17). For a brief moment, the swelling is halted, apparently by the thin outer membrane of the wall. This suddenly gives way, releasing the protoplasm, which rushes out pushing in front of it the grey material; this soon disappears, and one can then see a delicate spherical expanding membrane around the rapidly increasing mass of protoplasm which is pouring out of the mouth of the sporangium. Sometimes this mass appears at first almost spherical and homogeneous (Fig. 18), until about a third of the sporangial contents have escaped, after which its outline becomes irregular and delimitation of the spore units can be clearly seen; the second half, at least, of the outflowing protoplasm is divided up into distinct units, though these are very soft and become elongated as they emerge closely pressed

together in a mass in the middle of the sporangium and somewhat constricted as they pass through the opening (Fig. 19). Short flagella are usually visible on the distal periphery of the mass in the vesicle before discharge is complete, and soon elongate into long lashes which can be seen undulating in the clear region between the protoplasm and the membrane of the vesicle. At other times, the mass in the vesicle is lumpy and irregular in outline from the first, with deep cavities among the incompletely separated blocks of protoplasm, and the latter half or so of the emerging contents of the sporangium clearly consists of spores, clinging together closely. Most remarkable of all, flagella can be seen on them, long and active, waving as the spores move up to the mouth of the sporangium. They were present in the sporangium in figure 10, but are barely discernible in the picture. Their presence leaves no doubt as to the high degree of organisation reached by the spores while still within the sporangium. No previous record has been found by the writer, of the observation of flagella *within the sporangium* of any species of *Pythium*.

The irregular mass in the center of the vesicle shows at first an oscillatory motion due to the action of the flagella. Soon the separating units move individually, and take on a characteristic shape, with their somewhat flattened side, against which the vacuole lies, and from which the flagella arise, directed to the outside of the mass (Fig. 13, 20). The size of the vacuoles is comparable with that of the hyaline regions visible in the protoplasm before its discharge. They are contractile, and do not appear to take up neutral red, though smaller bodies in the zoospores, some of them apparently granular, have been seen colored with the stain. As the spores become separate and move apart, their movements become more and more pronounced until they are milling around in a frenzied manner, battering against the membrane of the vesicle. Sometimes, this membrane appears to disintegrate with the first rupture, allowing the zoospores to escape in all directions, but more often it is so tough that the first spore escapes through it with a jerk and the rest follow in its path, though some may remain swimming within the vesicle for a number of seconds, as though seeking a means of escape (Fig. 21). The torn remains of the vesicle are sometimes visible for a short time after the escape of the zoospores, but eventually disappear, though a minute fragment attached to the sporangium may remain visible for longer. It is easily possible to dissect off the mycelium a single sporangium bearing a vesicle containing swimming spores and to transfer it by means of a pipette to a drop of water on a slide, without causing any apparent injury!

In the first-formed sporangia in vigorous cultures, which go through an uninterrupted sequence of development and discharge, the papilla is sessile, and after its dissolution the mouth of the sporangium is seen to end abruptly with a smooth or sometimes reflexed rim (Fig. 35, 55). At other times, the papilla is formed at the apex of an outgrowth which remains after discharge as a short cylindrical collar (Fig. 35) or as a longer, sometimes irregular, exit tube (Fig. 34).

At temperatures between 18° and 25°C., the time from the first outrush of protoplasm to the escape of the zoospores from the vesicle is usually 14 or 15 minutes, but has been found to range from 12 to 17 minutes.

Less Characteristic Modes of Behaviour of the Sporangium

Under unfavorable conditions, when immediate discharge is prevented, the sporangium often rests for some time with a hemispherical region at the apex or on a short outgrowth entirely occupied by a clear substance apparently in a static condition, arrested in the early stages preceding sporulation (Fig. 27). If the spherical hyaline regions scattered through the protoplasm had already been formed, they remain with a hard, more refringent appearance than at first; and eventually a single large central "vacuole of senescence" (*vide* Drechsler, 1941) may develop (Fig. 54, a). If the water is refreshed, discharge of zoospores may occur in the usual manner after the disappearance of the large vacuole. More often, however, especially if the sporangium has been dormant for some time, one or more hyphae grow out. The extending hypha either carries with it the refractive material of the papilla or dissolves a path through it (Fig. 22, 23).

Certain less frequent irregularities in behaviour may be induced by more abnormal conditions, such as temperatures above 25°C., light from a bright microscope lamp, and the injudicious application of a coverslip. Even when the water is repeatedly refreshed, the presence of a coverslip for any but the shortest times is notably inhibitory to sporulation. Sometimes, for example, the sporangiophore swells terminally and then continues its growth as a hypha without cutting off a sporangium, illustrating the ready reversion of incipient sporangia to vegetative growth, which may occur at any stage in their development. This is shown in figure 53, in which the sporangial swellings at the tips of the branches resumed vegetative growth when the culture was disturbed for examination. Even the addition of fresh cool water may bring about a variety of irregularities in behaviour. In this way, premature discharge can be brought about, when the protoplasm emerges into the vesicle in an entirely homogeneous mass which never undergoes segmentation; or the protoplasm may pour out without the formation of a vesicle and segment into incompletely separated, irregularly shaped spores. Under some conditions, the spore units emerge sluggishly, becoming unusually elongated during their passage through the mouth of the sporangium. A discharged spore unit is then often seen to be drawn out behind into a fine thread by which it is attached to the spore unit following. Very occasionally, lumps of partially segmented protoplasm have been seen left behind in a partly empty sporangium.

These abnormalities in behaviour are significant only in so far as they reveal the possibility of misinterpretation, and indicate the importance of observing the sporangia under optimum conditions.

The Production of Secondary Sporangia

In a vigorous culture, proliferation through the empty sporangium soon occurs, the new sporangium being formed within (Fig. 37, 52) or more usually beyond (Fig. 40, 52) the previous one. In this process, the proliferating hypha may soften the septum and push it up with it (Fig. 55), as described in *P. proliferum* by Butler (1907), or it may dissolve a narrow channel through it (Fig. 40). It is not

uncommon to see as many as six sporangiophores emerging from the mouth of a single empty sporangium, and there are sometimes more; so many, in fact, that the sporangial opening may be completely blocked and some of the hyphae make their way out by boring through the side wall (Fig. 57). When this occurs, the tip of the hypha against the wall becomes very refractive; only a narrow passage is made, but the hypha expands again outside the wall. This continuation of the growth of the sporangiophore through an empty sporangium has been called "uniaxial replication" (1941, p. 480) and "uniaxial elongation" (1946, p. 855) by Drechsler, but the simpler term "internal proliferation" is preferable.

In cultures "where growth is vigorous, but the conditions are not such as favour zoospore formation" (Butler, 1907, p. 22), renewal takes place by subsporangial branching, or external proliferation, one or more secondary sporangiophores of variable length arising immediately below the septum of the primary one (Fig. 36, 38, 41).

This proliferation, internal and external, by several branches at the same time, for which the term "multiple proliferation" would seem appropriate, appears to be characteristic of *P. undulatum* alone. No other instance of its occurrence in the genus have been observed by the writer.

In addition to the above, secondary sporangia may also be formed by a type of branching showing no relation to the primary sporangium, or any constant order in the formation of the new ones (Fig. 53).

The Zoospore

The usual description of the *Pythium* zoospore as beanshaped, reniform or kidney-shaped seems to have been accepted without criticism by all writers on the subject, although a bean, and likewise a kidney, is a bilaterally compressed structure with a transverse groove in its ventral edge. On the other hand, a swimming spore of *P. undulatum*, which may be taken as typical of the genus as a whole, is dorso-ventrally compressed with a longitudinal spirally-twisted groove in its ventral surface. This has been well illustrated by Matthews (1932, plate 12, fig. 4). It is this groove which causes the pointed appearance, as it is especially pronounced at one side of the anterior end, and flattens out on the opposite side at the posterior end. An impression of a bean-like shape is gained only from an end-on view of the spore, when the longitudinal groove is seen in profile as an indentation on the ventral side. With changes in the level of focus, this indentation varies about a median position, on account of the spiral twist of the groove. This is not the usual view of a free-swimming spore, but is the one usually presented by a spore that is about to settle down and encyst, and is frequently seen on the periphery of a mass of spores maturing in a vesicle, when the groove is directed towards the outside of the mass, and the two flagella arising out of it, in a median position, can be seen undulating in the region between the protoplasm and the membrane of the vesicle. At this time, the spores may seem to have a less definite shape than they do actually possess, owing to their twisting and writhing motion, and to their irregularity of outline, due to the presence of the groove. Although they are soft and compressible, a fact which may be readily

noted as they squeeze through the narrow opening of a tough vesicle, the shape of free-swimming zoospores is a definite and characteristic one.

The zoospore swims in a clockwise (as seen under the microscope) spiral path, rotating on its long axis at the same time. Whether the twist of the groove takes effect in the rotation or in the wider spiralling of the forward movement, or in both, is difficult to determine. The longer flagellum is directed posteriorly, the shorter one anteriorly. Zoospores swim for a variable time; in a drop of distilled water on a slide, they may settle down almost at once, or they may swim for half an hour or more. Just before settling, a spore sometimes makes faster, jerky movements along short paths in various directions, and then suddenly stops. It settles on its pointed anterior end, presenting an "end-on" view, and vibrating slightly as it becomes spherical, until finally it is quite motionless within a delicate wall. One or two slightly thickened twisted threads may be visible at one side for a short time afterwards, representing the remains of the flagella. The newly-encysted zoospore is 13–15μ in diameter, an unusually large size for the genus. Since germination involves the swelling of the encysted spore, it is important that measurements recorded are of *newly-encysted* spores. The prompt encystment of swimming spores can be promoted by transferring them in a pipette to a drop of fresh water on a slide and applying a coverslip. In their large size, as well as in their subsequent behaviour, the zoospores of *P. undulatum* resemble those of *P. palingenes* (Drechsler, 1941).

The Behaviour of the Zoospore after Encystment

Germination.—In a nutritious medium, the encysted zoospore produces a germ hypha which arises as a blunt, rounded, hyaline swelling (Fig. 42). Soon a few granules and droplets appear in the extending hypha, and when it has reached some length the protoplasm has the usual coarse granular appearance, except at the tip, which is finely granular or hyaline. When the cyst wall is very thin, it appears to extend with the growing hypha, but when some thickening has occurred the germ hypha is constricted at the base where it passes through the thickening. While the hypha is extending, a large central vacuole develops. This is not comparable with the contractile vacuole of the swimming spore. It accumulates neutral red readily from a dilute solution, while the contractile vacuole does not. However, when the sporeling is well established, as for example in an agar film slide culture, protoplasm may pass back into the cyst, which then loses the vacuole and may possibly serve as a reservoir for the protoplasm. From the germ hypha an extensive branched mycelium develops (Fig. 43–46). The sensitivity of germ hyphae to aeration and food concentration has been observed. Spores encysted under a coverslip, near the edge, have often been found with their germ hyphae lying more or less parallel in a row, directed towards the edge of the coverslip. Also, scattered oil drops liberated from the hemp seed substratum are sometimes reached by a number of germ hyphae from all directions, from encysted spores round about.

Under less favorable conditions, a miniature sporangium is produced at the tip of a hypha in which septa are successively formed as the protoplasm draws

out of the basal parts (Fig. 49–51). The sporangium is papillate and appears in all ways but size similar to the sporangia formed on the ordinary mycelium. Discharge has not been observed by the writer, but several empty sporangia have been seen.

Repeated emergence.—Spores in sterile distilled water on a slide have been observed to liberate zoospores like themselves through a short exit tube. The empty cysts with their exit tubes are frequently seen (Fig. 47). The addition of fresh water to a slide bearing zoospores which have begun to germinate by a hypha will sometimes cause them to retract the protoplasm from the hypha and discharge a zoospore through an exit tube (Fig. 48). The liberation by *Pythium* zoospores of secondary spores like themselves was described in 1872 by Cornu, but without a name for the phenomenon, and Butler believed himself to be the first to report it, in describing *P. diacarpum* in 1907. He called it "diplanetism", and this term has been used over and over again, despite its entirely different meaning in the Saprolegniaceae, in which it was first applied. Variations on this term have been supplied by Drechsler, who included *Pythium* in his discussion on the "repetitional diplanetism in the genus *Phytophthora*", and he has also used a number of other terms. Among these are "an iterated swarming stage" (*P. palingenes*, 1941) and "repetitional development" (1946). The same phenomenon in *Dictyuchus* was described for the first time by Weston (1919) as "repeated emergence", and this name has been adopted for *Pythium* by Matthews and for *Phytophthora* by Blackwell (1949). Middleton also uses it, but in addition describes the phenomenon as "repetitional emergence or secondary spore formation."

OTHER OBSERVATIONS ON *PYTHIUM UNDULATUM*

The fungus has been grown on a variety of media at various temperatures; some casual examinations of waterlily leaves and petioles have been made for possible intramatrical oogonia; all possible combinations in pairs of the three isolates in the same dish have been tried, in order to test for heterothallism; but there has never been any sign of the formation of oogonia by any of the isolates. However, thickwalled chlamydospores are formed abundantly, with a structure and behaviour agreeing closely with those figured and described in detail by Drechsler (1946). Drechsler's conviction that they are parthenospores homologous with the oospores of *P. helicoides*, stated in his summary, is revealed further by his inclusion of *Pythium undulatum* in a paper entitled: "Several species of *Pythium* peculiar in their sexual development."

The invasion of the hyphae of *P. undulatum* by those of three other species of the same genus when grown in opposition to it is described by Drechsler. He does not record, however, the passage through them of other hyphae of its own. Such "intrahyphal hyphae" (Buller, 1933) have been seen frequently on cornmeal agar, supposedly due to some unfavorable condition, perhaps osmotic pressure, in the agar.

SOME OBSERVATIONS ON *PYTHIUM PROLIFERUM* DE BARY

It would appear from the literature that a number of different strains, at least, have been described under the name *Pythium proliferum*. This may account in

part for the differences in the records of the behaviour of this species. Cultures of a strain from Florida, measurements and other characteristics of which are recorded in a previous paper (Goldie-Smith, 1951), were treated according to the method described above, for comparison with *P. undulatum*. Three stages of discharge are shown in figures 3 to 5. As described by Butler (1907), one or more quite large vacuoles, sometimes irregular in outline, are usually present and disappear just before discharge; but there is an additional vacuolation which, with the very conspicuous lumping of the protoplasm also observed by Butler, makes the sporangia which are just ripe for discharge easily detectable even under low magnification, though so much smaller than the sporangia of *P. undulatum*. They can therefore be watched and timed without the use of a coverslip. The outflow is much faster in *P. proliferum*, but the maturation time is about the same as in *P. undulatum*, at comparable temperatures. This time has been found to vary between 11 and 15 minutes at 25°C. and between 7 and 9 minutes at 33°C. At 33°C., too high a temperature even to look for healthy behaviour in *P. undulatum*, normal spores of *P. proliferum* are formed which swim away in the usual manner, illustrating the greater range of tolerance of this species, already apparent in its prolific growth under a variety of conditions and the relative ease with which the formation of large numbers of active sporangia can be induced.

Discussion

The differentiation within the sporangium.—Sporangial germination in *Pythium* is generally described as characterized by the passage of the protoplasm "in an undifferentiated state" into an "evanescent vesicle" (Matthews, 1931; Middleton, 1943), despite the fact that various degrees of differentiation before discharge are reported in the earlier literature. The situation is perhaps better expressed by Butler (1907) as follows: ". . . the genus *Pythium* is separated from all the rest" (of the Peronosporaceae) "by liberating its zoospores in an imperfectly differentiated state into a bladder at the mouth of the sporange, in which differentiation is completed."

"Transitory beginnings of division" in the sporangium were recognized by de Bary (1887), who saw light spots separated by darker, dense granular lines seeming to indicate division. Büsgen (1882) believed the protoplast of *P. proliferum* at this stage to be divided up by a network of groups of granules in rows, which he interpreted as cell plates. The network soon disappeared, leaving equidistant vacuoles and bright spots, which were frequently grouped around a specially large vacuole. In *P. debaryanum*, Hesse (1874) observed circular areas separated by *light* streaks which seemed to foreshadow a separation but disappeared as quickly as they came. Butler was unable to agree with Büsgen and Hesse, and in a general account of sporulation in *Pythium* reported seeing nothing comparable with Büsgen's "Spuren eines Körnernetzes" or with Hesse's "helle Grenzstreifen," but only "sharp dark dots surrounded by a clear area" which he considered might be nuclei. Butler noted, however, that the protoplasm became "lumped into heaps around the periphery of the sporangium," and that this lumping was more difficult to see sometimes than others. Furthermore, de-

scribing *P. rostratum*, Butler stated his belief that the spore-origins are definitely formed before discharge, and "though fused into a mass in which individual spores cannot be made out, yet each nucleus has obtained a hold on a certain mass of cytoplasm which passes out with that nucleus."

The genus was founded by Pringsheim (1858) on *P. monospermum* (and a species which was later transferred to the genus *Lagenidium*), and it appears that in such species with filamentous sporangia the protoplasm is indeed extruded in a less differentiated condition than in species with spherical sporangia. The ejected protoplasm of *P. adhaerens* is described by Sparrow (1931, b) as a "smoothly granular, homogeneous mass." A similar condition would seem to be implied by Saksena's (1935) figures and description of *P. deliense*. These species vary, however, from time to time, for the occasional cleavage of the sporangial contents before emergence has been described in *P. gracile* by Büsgen, *P. monospermum* (*P. fecundum*) by Wahrlich (1887), and in *P. aphanidermatum* (*Rheosporangium aphanidermatus*) by Edson (1915). Variation in the behaviour of different sporangia of the same species was observed as early as 1872 by Cornu, who noticed that while the behaviour of *P. proliferum* was sometimes similar to that of *P. monospermum*, at other times the zoospores were completely formed within the sporangium. In the same species, Marshall Ward (1883) saw transitory segmentation in a single sporangium, and Höhnk (1933) reported an incomplete segmentation within the sporangium as the usual process, although it was generally followed by a homogeneous phase as the protoplasm flowed out.

Correlated with this variation in the accounts of the process of differentiation is the lack of agreement among the different investigators as to the time required for the maturation of the zoospores within the vesicle. For *P. proliferum*, among the times which have been reported are: 1 or 2 to 30 mins. (Cornu, 1872); about 20 mins., sometimes more than 2 hours (Höhnk, 1933); 16 mins. (de Bary, 1860); and 16, on one occasion only 13 mins. (Marshall Ward, 1883). Among species with filamentous sporangia, the following times have been reported: *P. aphanidermatum*, usually 30 mins. (Edson, 1915); *P. afertile*, less than 30 mins. (Kanouse and Humphrey, 1927); *P. deliense*, 20 to 25 mins. (Saksena, 1935); and *P. gracile*, about 23 mins. (Marshall Ward, 1883). While it seems most likely that this time depends largely on the degree of differentiation previously reached, it is possibly also determined by the effect of environmental conditions, temperature in particular, on the *rate* of the organization process. It is therefore clear that it is important to describe the process under optimum conditions, differing in different species, and to refrain from over-emphasizing an odd sporangium or two showing anomalous behaviour. For this reason, care has been taken in this study to observe the sporangia "in the first flush of growth under good conditions" (Blackwell, 1949), and to describe the characteristic or usual type of behaviour.

Vacuolation.—Vacuoles play an important part in the process of sporulation Butler (1907) says: "A ripe sporangium ready to discharge is always vacuolated. One or more vacuoles may occur, and their size is variable, small usually in *P. intermedium* and *P. vexans*, larger in *P. rostratum* and *P. proliferum*." The vacu-

oles of *P. proliferum* were also observed by Büsgen, Marshall Ward, and Höhnk (1933). Vacuoles are reported in the filamentous sporangia of *P. monospermum* by Wahrlich, *P. gracile* by Butler, *P. aphanidermatum* by Edson, and *P. adhaerens* by Sparrow (1931, b). In addition to those present in the hyphae and young sporangia, it seems that several kinds of vacuoles, with different functions, may be associated with the mature sporangium, though it is not always clear in previous accounts which kind is implied. The following three types can be distinguished: relatively large, irregular vacuoles which swell and disappear immediately before discharge; small, regular, equidistant vacuoles which also, according to most accounts, disappear before discharge; and a large central vacuole which ultimately develops in resting sporangia, aptly called by Drechsler (1941) a "vacuole of senescence." Perhaps a fourth should also be included, the "subapicular vacuole" described by Butler in the development of the papilla. While this and the first two types are directly concerned in sporulation, the vacuole of senescence is opposed to it. After its formation in some species, sporulation can no longer take place, and the only further development possible is direct germination by means of a hypha. Under suitable conditions, in other species such as *P. undulatum*, the sporangium may undergo a process of rejuvenescence, so to speak, when the large vacuole of senescence disappears with the initiation of the normal process of sporulation. In *P. undulatum*, as in *P. epigynum* (Höhnk, 1932), the longer the sporangium has been in the resting state, the smaller are the chances of its undergoing sporulation when the conditions are changed.

The greatest importance was attached by Butler to the first kind, of which the significance had previously been overlooked. He believed the sudden enlargement of the vacuole to be due, as in the Saprolegniaceae, to the inflow of water after the rupture of "the protoplasmic investment between the spore origins" resulting from the lumping of the protoplasm. Its subsequent collapse and disappearance after reaching maximum pressure is accompanied by a considerable decrease in the size of the sporangium, part of its contents, in Butler's belief, passing out through the sporangial wall, and part being taken up by the spore origins which consequently swell and lose their distinctness of outline, giving the effect of a "homogeneous stage." With this interpretation, organization is considered well advanced by the time that the protoplasm emerges into the vesicle, where the process described by Butler as the "*final fashioning*" of the zoospores occurs, "corresponding to the fourth stage in the *Saprolegniaceae*."

In *P. undulatum*, the preliminary vacuolation seems to be relatively inconspicuous and very rapid. It is presumably represented by the cracks and fissures that develop throughout the protoplasm immediately before discharge, but seeming to be the cause rather than the result of the separation of the spore origins. Although these have been seen lightly stained with neutral red, difficulty in getting them to acquire any depth of color is undoubtedly related to their dynamic state and special function. The same is probably true of the equidistant spherical vacuoles. The vacuole or vacuoles of senescence, on the contrary, which may come to occupy more than a third of the volume of the sporangium, readily accumulates neutral red and acquires a good color.

In one species described by Drechsler (1941), *P. marsipium*, the preliminary vacuolation process is either absent or unusually inconspicuous. Drechsler writes: "During the period immediately preceding discharge, the sporangium here does not reveal any vacuole comparable with the large lacuna-like vacuole that, in most congeneric forms, increases in size until it abruptly disappears very shortly before the protoplasmic materials are expelled." The "numerous, more or less evenly distributed, subspherical hyaline bodies, approximately 3μ in diameter" may possibly represent the vacuoles of the second type described above. A lumping of the protoplasm about them results in an appearance suggestive of cleavage comparable, in Drechsler's opinion, with that of *Phytophthora* sporangia. He writes: "Manifestly, at the time of their discharge, the sporangial contents here have already undergone the initial stages of reorganization that elsewhere are accomplished during the first 5 minutes of their sojourn within the vesicle. Since the further stages of transformation follow in the sequence usual for species of *Pythium*, the laterally biciliate zoospores are ready to be liberated after 15 to 20 minutes, rather than after 20 to 25 minutes as in most congeneric forms." *P. marsipium* thus somewhat resembles *P. undulatum* in behaviour. The maturation time, 15 minutes or less in *P. undulatum*, is even shorter than in *P. marsipium*. Drechsler's statement that the usual time in "congeneric forms" is 20 to 25 minutes needs substantiation. As mentioned under the previous heading, it seems that differentiation tends to proceed further in spheroid than in filamentous ones, with a shorter maturation time as a consequence, and a truer picture would perhaps be gained by averaging the times for the two types separately.

The vacuolation process may possibly also assist in the ejection of the sporangial contents. Butler believed the protoplasm to be "attracted out by the formation of a chemotactic substance in the subapicular vacuole." The force of positive aerotaxy which Hartog believed to operate in *Saprolegnia*, and possibly in *Phytophthora*, seems unlikely in *Pythium*, in view of the confining effect of the vesicle, although his alternative theory of negative pneumotaxy is more acceptable since the protoplasm may well tend to escape from the products of its own metabolism. Further, the outflow might be a result of the decrease in pressure at the mouth of the sporangium, caused by the expansion of the vesicle. However, considering the rapidity of the process, and the completeness of the evacuation, some positive force seems more probable. The protoplasm may possibly be pushed out of the sporangium by the pressure of the fluid in the vacuoles, as suggested by Marshall Ward, or by the swelling of the wall of the sporangium, or of some substance derived from it.

Relations with Phytophthora.—Variation in behaviour in response to different conditions may be the explanation of the confusion between *Pythium undulatum* Petersen and *Pythiomorpha undulata* Apinis. This problem has been discussed by Drechsler (1946). While Apinis believed he had the same fungus as Petersen, which he transferred in 1929 from the genus *Pythium* to *Pythiomorpha*, in subsequent records the same specific epithet has been applied to the two genera. Thus, *P. undulatum* has been recorded by Dissmann (1927), de Wildeman (1931),

Matthews (1931), Sparrow (1932), Lund (1934), Drechsler (1941, 1946), and van Beverwijk (1948). *P. undulata* has been recorded by Cejp (1932) and Lund (1934).[2] Lund thus claimed to have seen both. His detailed description of *P. undulata* agrees closely with that of Apinis, and includes the definite statement that the zoospores are formed in the sporangium and escape through a terminal opening. His record of *P. undulatum*, on the other hand, is based on the observation of a single sporangium which formed a vesicle. The genus *Pythiomorpha* has been proved invalid (Blackwell, Waterhouse and Thompson, 1941), and it may be that there is in existence an aquatic species of *Phytophthora* resembling *Pythium undulatum* in all features except sporulation. It is much more likely, though, that the two descriptions are referable, as Apinis himself supposed, to a single organism. This view is strengthened by the facts that both Apinis and Lund described the occasional encystment and germination of the spores within the sporangium, and that Lund also reported direct germination of the sporangium by hyphae, both indicating that some of the observations at least were made under conditions promoting ab- or sub-normal behaviour of the sporangia.

Discharge of *P. undulatum* without the formation of a vesicle is described as rare by Dissmann, and considered as an irregularity. He commented that the slow rate of emergence of the spores and the irregularity of their form indicated that they were of doubtful vitality. Drechsler has suggested that *Phytophthora* sporangia may have been present in some of Dissmann's cultures, figured with wide mouths and no exit tubes, some of them nested one inside another as a result of internal proliferation in such a way that there would appear to be no room for the formation of a vesicle. This suggestion is not justified, for the present study has shown that the first-formed sporangia in a vigorous culture usually discharge with a sessile papilla, without the formation of an exit tube (Fig. 21, 55), and also that nesting of the type figured by Dissmann does indeed occur in pure cultures of *Pythium undulatum* (Fig. 35, 58).

We are thus led to conclude with Sparrow (1932) that *Pythiomorpha undulata* is synonymous with *Pythium undulatum*. However, the confusion is an interesting reflection of the difficulty that is met with from time to time in distinguishing between members of the two genera *Pythium* and *Phytophthora*. In 1910, describing *Pythium palmivorum*, Butler stated his belief that the formation of a "thin gelatinous vesicle into which the protoplasm passes in a uniform granular mass" is "practically universal in the genus *Pythium*" and "the only absolute mark of distinction of this genus from *Phytophthora*." In the same fungus, however, particularly during the cooler months of the year, a complete or nearly complete segmentation into zoospores within the sporangium also occurred, and in 1919 it was renamed by Butler *Phytophthora palmivora*. In 1938, Shanor de-

[2] An interesting record was given under the name *Saprolegnia elongata* by Massee in 1891 (British Fungi, p. 217). As Coker (1923, p. 76) has pointed out, this is apparently "based on mixed material, in part a *Pythium* and in part a *Saprolegnia*." The sporangia in plate 3, figure 47, closely resemble those of *P. undulatum*. The sporangium is described by Massee as "terminal, elliptical, 50–60 × 18–21μ, filled with zoospores which escape from an opening at the apex." The sporangiophores are undulating, with a conspicuous widening under the sporangium, and proliferation is shown.

scribed under the name *Phytophthora stellata* a *Pythium*-like fungus in which the process of sporulation is said to resemble that of *P. palmivora* and sometimes very closely approaches that usually found in the genus *Pythium*. He writes: "Forms in which the sporangial contents have been freed into a vesicle entirely undifferentiated and forms in which the zoospores are liberated into the vesicle almost completely formed have been observed." Similar variations in behaviour are reported for *Phytophthora parasitica* by Dastur (1913) and Rosenbaum (1917). Among other records of *Phytophthora* species occasionally forming a vesicle are *P. cactorum* (Rosenbaum, 1917; Wormald, 1919), *P. arecae* (Rosenbaum, 1917), *P. colocasiae* (Blackwell and Waterhouse, 1930), and *P. cryptogea* (Blackwell, 1949). It appears, however, that the vesicle formed in these instances is usually truly evanescent in comparison with the more durable one of *Pythium* species, and the formation of a "relatively tough" one is rare (Blackwell, 1949). The influence of environmental conditions is evident in these accounts, although due consideration has not always been given to it, and emphasis has often been on the aberrant rather than on the normal. The importance of "suitable environmental conditions" is stressed by Middleton (1943, p. 19). "When properly treated," he writes, "the sporangia germinate in a characteristic and peculiar manner." Despite the criticism of Fitzpatrick (1923) and Sparrow (1931, c), who favored the merging of the two genera *Pythium* and *Phytophthora*, the belief is stated by Middleton that the mode of zoospore production "may be retained as a criterion of generic segregation . . . if its value and limitations are thoroughly understood." All the same, there is need for qualification in the statement on page 4 of his monograph: "In no instance are zoospores formed within the sporangium." It would be more accurate to say instead: "In no species of *Pythium* under optimum conditions are zoospores *completely* formed within the sporangium."

Other features which have been cited as points of distinction between the two genera are the growth habit, the branching of the sporangiophore, the width of the sporangial openings, the formation of an exit tube, and the structure of the hyphal wall. Drechsler (1946) describes the mycelium of *P. undulatum* as offering "the generally flexuous appearance familiar among species of *Pythium* rather than the stiffly branching aspect common in species of *Phytophthora*," and the rightangled branching of *Phytophthora* species, frequently with a constriction at the base of the branch, has been pointed out by Miss Blackwell (1949). Middleton's statement that "sporangiophore branching is unusual in *Pythium* and not uncommon in *Phytophthora*" is difficult to interpret, for he does not define the limits of a sporangiophore, and it is not clear whether he has taken into account the internal proliferation and subsporangial branching (external proliferation) that occur abundantly in several species of *Pythium* with spheroid sporangia, among them *P. undulatum* and *P. proliferum*. In these two species, branches also arise in various positions on the sporangiophores with no relation to existing sporangia. While it is true that the sporangial openings of *Pythium* are usually narrow in comparison with the "generous" ones of *Phytophthora* (Drechsler, 1941, p. 850), and that an exit tube is usually formed, in a number of species of

Pythium, such as *P. helicoides*, *P. palingenes*, and *P. undulatum*, the papilla may be sessile. All the above are rather ill-defined and somewhat flexible criteria of distinction between *Pythium* and *Phytophthora*, but are nevertheless convenient and generally reliable when considered together. A more definite one will be found if further evidence is supplied in support of Thomas's (1942, 1943) finding that the hyphal wall of *Phytophthora* species differs in structure from that of species of *Pythium*. The structure of the wall has not been investigated in *Pythium undulatum*, and the reaction with chlor-iodide of zinc is here not considered of generic significance.

Summary

While *Pythium undulatum* certainly falls within the limits of the genus *Pythium*, it has some features which seem to place it on the edge of the group, bordering on the genus *Phytophthora*. Chief of these is the high degree of organization reached before discharge by the sporangial contents, which sometimes emerge already segmented into flagellated zoospores. The observation of flagella on the spores while still in the sporangium has not previously been reported in any species of *Pythium*. Under normal conditions discharge always occurs into a characteristically tough vesicle. In the type of vacuolation undergone just before discharge, this species resembles *P. marsipium* Drechsler. A survey of the literature reveals previous reports of varying amounts of differentiation within the sporangium of species of *Pythium*, and shows the need for a revision of the general concept of the process in the genus. The familiar description of the newly-discharged sporangial contents as "undifferentiated" is not applicable to all, if to any species. Comparison of the shape of the *Pythium* zoospore with that of a bean or kidney is also a misconception. Further study of a wide range of species including both spheroidal and filamentous types is necessary, but must be carried out under optimum conditions, and due consideration must be given normal, as distinct from abnormal, behaviour. *P. undulatum* responds with outstanding sensitivity to environmental factors, in both form and behaviour, and its wide variation probably accounts for the discrepancies in earlier descriptions, and for the confusion between *Pythium undulatum* Petersen and *Pythiomorpha undulata* Apinis, which are almost undoubtedly a single organism. A form of dimorphism is shown, in that the first-formed sporangia in a vigorous culture are large and ellipsoidal, and borne on wide undulating sporangiophores very distinct from the narrower hyphae of the later growth on which smaller sporangia develop; the latter are frequently ovoid or obpyriform in shape, thus differing from the earlier ones in proportions as well as in size. A further unique characteristic of this species is the frequent renewal of the sporangia by multiple internal or external proliferation.

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EXPLANATION OF PLATES

PLATE 24

Pythium proliferum de Bary. ×360

FIG. 3. The first gush of protoplasm into the expanding vesicle.

FIG. 4. The emergence of the partly differentiated protoplast.

FIG. 5. Maturation within the vesicle.

Pythium undulatum Petersen

FIG. 6. Sporangium showing a refractive apex and equidistant vacuoles. ×360.

FIG. 7. The lumping of the protoplasm. ×250.

FIG. 8–9. The first outrush of the protoplasm. Fig. 8. ×215. Fig. 9 ×516.

FIG. 10. The emergence of spores at an advanced stage of differentiation. (Note: these spores actually bore flagella visible while still within the sporangium!)

FIG. 11–12. Successive pictures of the same sporangium just before discharge, at different levels of focus. ×560.

FIG. 13. Spores maturing within a vesicle. ×360.

PLATE 25

P. undulatum Petersen

FIG. 14–21. Stages in the development and maturation of the zoospores. ×330.

FIG. 22–33. Common but less typical sporangial forms. ×425. Fig. 22–23. Continued growth by means of a hypha. Fig. 24–33. Various shapes and sizes of sporangia.

FIG. 34. A sporangium that had formed an apical protrusion and exit tube. ×425.

FIG. 35. Nesting. ×425.

FIG. 36, 38. Subsporangial branching. ×175.

FIG. 37. Internal proliferation. ×175.

FIG. 39. Sporangiophore closely spiralling and undulating. ×175.

FIG. 40–41. Sporangiophores borne on stolon-like hyphae from which narrow vegetative hyphae are also arising. ×175. Fig. 40. Multiple internal proliferation. Fig. 41. Subsporangial branching.

FIG. 42–51. Germinating spores. ×340. Fig. 42–46. Direct germination by a hypha. Fig. 47. Cyst, with exit tube, after the re-emergence of the zoospore. Fig. 48. Spore which had withdrawn its contents from the germ hypha and undergone repeated emergence, as a result of a change in conditions. Fig. 49–51. The formation of a miniature sporangium.

PLATE 26

Pythium undulatum Petersen

FIG. 52. Internal proliferation. ×175.

FIG. 53. Branched sporangiophore. ×120.

FIG. 54. Small sporangia of the ovoid type; a, with a central vacuole of senescence; b, immediately after discharge. ×80.

FIG. 55–57. Internal proliferation by hyphae. ×420. Fig. 55. By a single hypha. Fig. 56. By several hyphae. Fig. 57. By very many hyphae, four of which are seen passing through the sporangial wall, at a, b, c, and d.

FIG. 58. Nesting. × 350.

PLATE 24

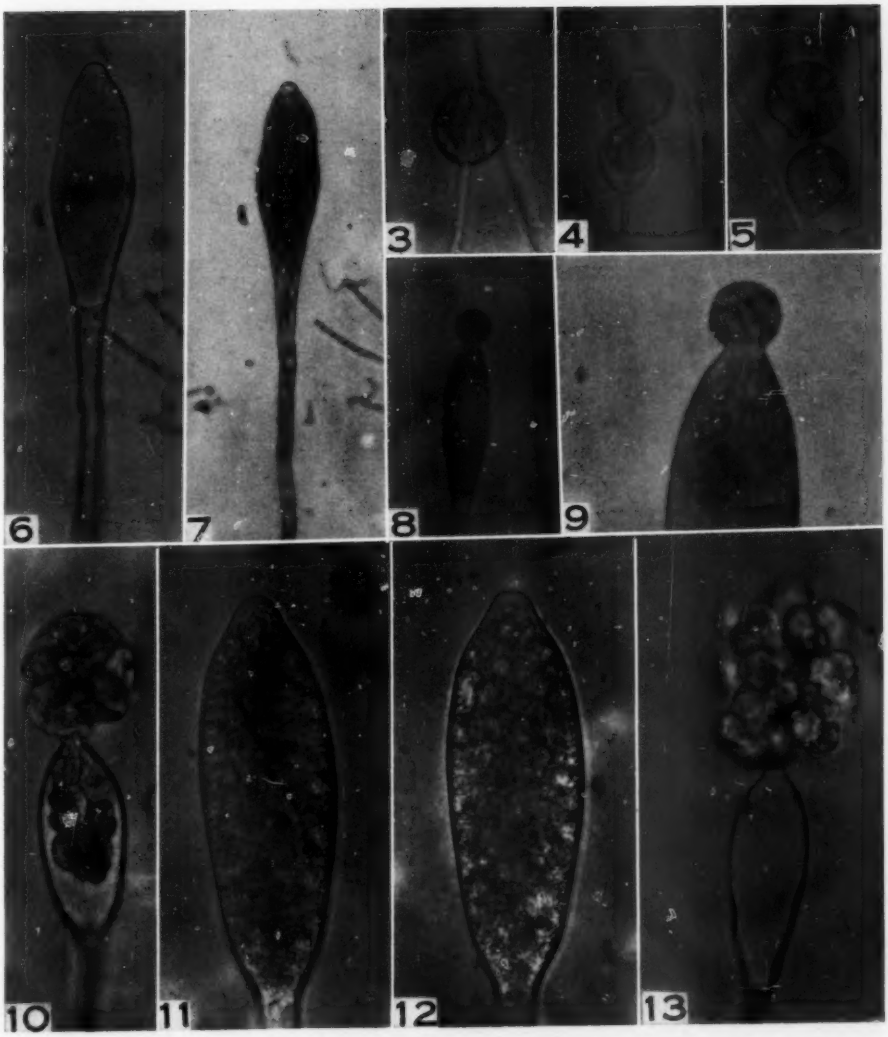
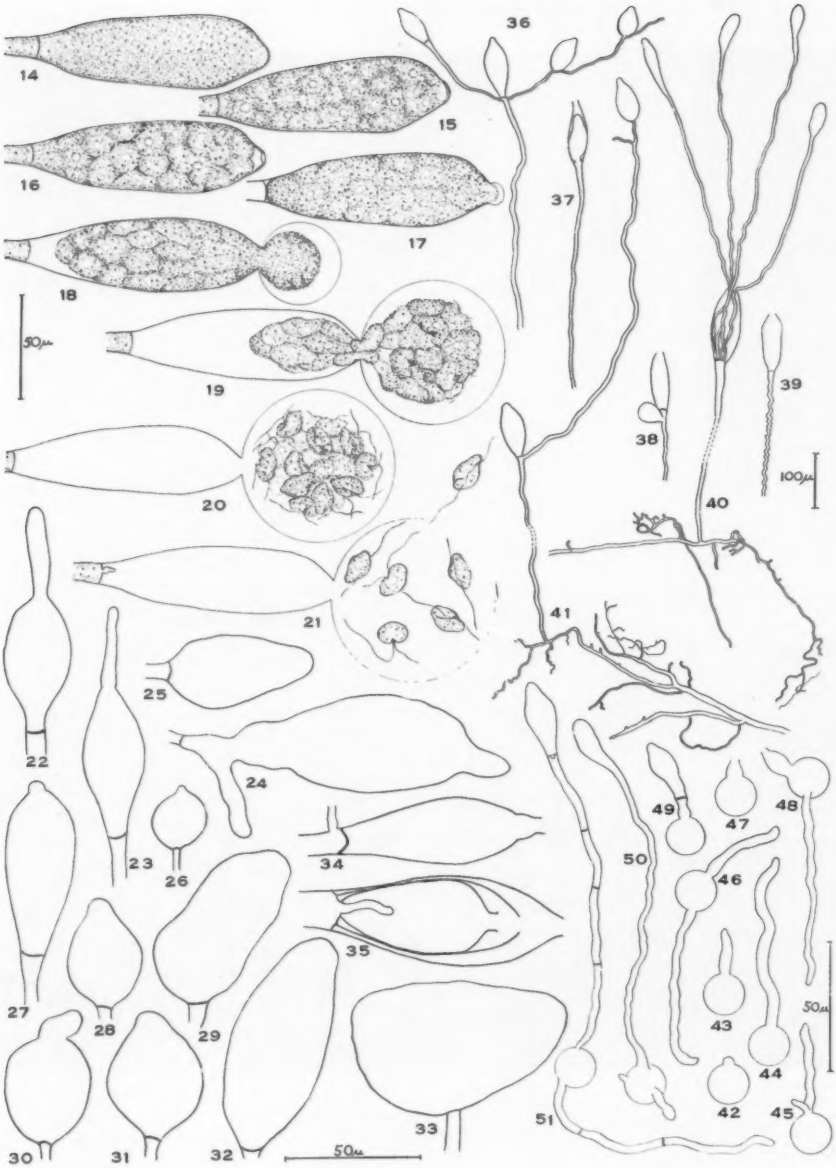


PLATE 25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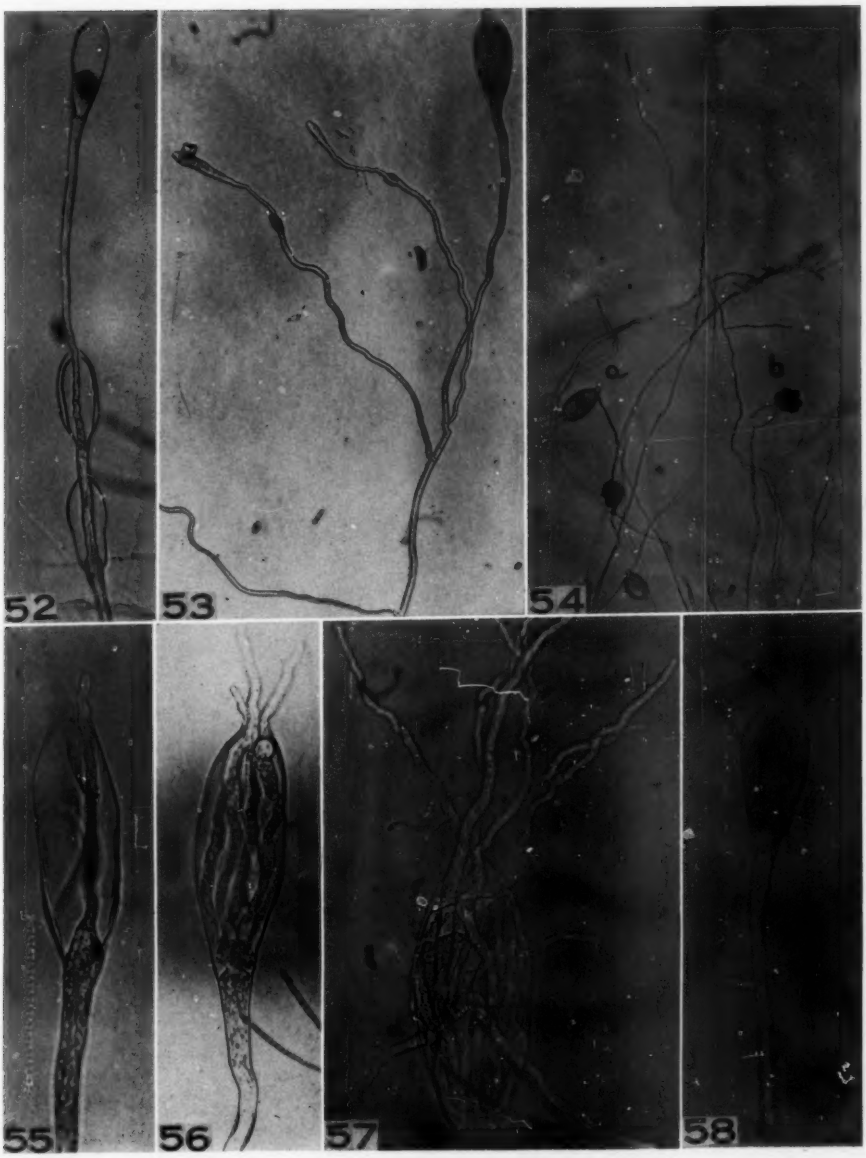
PLATE 26